VOLUME XLII NUMBER 4

BOTANICAL GAZETTE

OCTOBER, 1906

THE DEVELOPMENT OF AGARICUS CAMPESTRIS.*

GEO. F. ATKINSON.

(WITH PLATES VII–XII)

IN some respects the history of the study of the Hymeniales does not present the same progress which can be seen in the other groups of fungi, or indeed in nearly all other groups of plants. The earliest period, that of the study and classification of species and genera, presents in the main the same aspects which have been characteristic of the early study of all plants; but the progress made up to the present time is not in proportion to the time and energy expended, due to certain difficulties, some inherent in the nature of the plants themselves, and others due to the lack of an adequate knowledge of their anatomy and development.

The second period, that of the study of the morphology and development, began more than half a century ago. It is true that in the early part of the 19th century, nearly a century ago, quite an elaborate theory of the development of the Hymeniales, especially the Agaricaceae, was evolved by NEES VON ESENBECK.[1] But his theory, embellished as it is with his philosophical ideas of the evolution and metamorphosis of these plants from the puffballs and truffles; in which he was evidently influenced by the philosophy expressed in the *Vorwort* of GOETHE'S *Farbenlehre*, that in a book dealing with

* Contribution from the Department of Botany of Cornell University. No. 110. Paper presented before Section G, of the A. A. A. S., at the New Orleans meeting.

[1] Das System der Pilze und Schwämme, ein Versuch von Dr. C. O. NEES VON ESENBECK, I–XXXVI, 1–329, 44 plates, Würzburg, 1816. See also NEES VON ESENBECK, Plantarum Mycetoidarum in Hort. Bonn. obs. evolutio. Nov. Act. Natur. Curios. **16**: pars 1. 1832, for development of *Agaricus volvaceus*.

natural phenomena the writer should make use of a lively imagination in order to make it real to the reader; and especially because there is such lack of definiteness as to the forms studied, though it is quite evident he refers more especially to species of Amanita, presents little that is helpful to the present discussion.

At that early period it was an important forward step to show, as DUTROCHET[2] did in 1834, that the large fungi were only the fruit bodies of the plants then known as "Byssus," which spread usually underground or in the substance of organic bodies; and for TROG[3] in 1837 to recognize the two different parts in the life history, the vegetative stage or mycelium and the fruiting stage or carpophore, and that this is the product of germinating spores; though MICHELI had stated as early as 1729[4] that the fruit bodies of some fungi did not come immediately from the seed (spores), but the seeds first produce a large root which grows for several years in the ground, and then gives rise to the fruit body (referring to *Polyporus tuberaster*). But during the early and middle portion of the 19th century the work on the morphology and anatomy of these plants, and the descriptions and illustrations of species, was far in advance of the work on development and the organization of the parts of the fruit body. Unfortunately the study of the morphology and development of the Hymeniales has not kept pace with the same studies in other groups of plants. J. SCHMITZ[5] studied the very early stages of five different species. While the work appears to be very carefully done for that early time, it does not meet modern requirements; and while his results perhaps in the main are not far out of the way for the species studied (*Cantharellus sinuosus*, *C. tubaeformis*, *Cortinarius bulliardi* (Pers.) Fr., *Coprinus niveus*, *Hydnum imbricatum*), it will be seen later he was led into a mistake in formulating a general law, based on these five species, to apply to the development of all the pileate fungi.

[2] Mém. **2**:173. 1834.

[3] Ueber das Wachsthum der Schwämme, Flora **20**:609. 1837.

[4] Nova plantarum genera 134. 1729.

[5] Mycologische Beobachtungen, als Beiträge zur Lebens- und Entwickelungsgeschichte einiger Schwämme aus der Klasse der Gastromyceten und Hymenomyceten. Linnaea **16**:141–215. *pls. 6–7.* 1842. See especially the part II, Ueber die Bildung neuer Theile bei den Hymenomyceten, vorzugsweise den Pileaten. Idem 168–179.

BONORDEN[6] deals briefly with the anatomical structure of the genera recognized by FRIES in his *Epicrisis*. He does not discuss the differentiation of the parts in the young fruit body, but describes somewhat in detail the different forms of the universal veil, its mode of dehiscence, and its relation to the partial veil and the pileus in certain species of Lepiota (pp. 178–181). He says, very briefly (p. 8): "From the mycelium rises the fruit body of the fungus (stroma, thallus), either naked from the first or enclosed in an envelop (velum, volva). The latter consists always of very much elongated cells, tubes, which are like the tissue of the mycelium, and has therefore always a structure very different from that of the fruit body of the fungus; it is to be regarded as a continuation of the mycelium. The envelop is ruptured by the further growth of the pileus and is thrown off, but sometimes remains a part of the same and forms the epidermis of the pileus, on which account this is so different in structure in the case of the gastromycetes and pileated fungi from the other parts of the fungus."

H. HOFFMAN contributed at that time some important work on the anatomy and morphology, and as early as 1856 gave a very brief account of the origin of the hymenium of *Agaricus campestris*.[7] In speaking of his studies of the developmental history of the lamellae in very different types (*l. c.*, p. 145) he cites three extremes: *Agaricus carneo-tomentosus* (*Panus torulosus*), where they arise at the apex of the young fruit body; *Agaricus campestris*, where they originate deeper in the interior and develop laterally; and *Hymenogaster klotzschii*, where they remain concealed in the interior of the fungus. In describing the development of *Agaricus campestris* (p. 145) he says (I give a free translation): "It begins, as BULLIARD has already very well represented (Champ. d. France. *pl. 514, fig. L.* 1791–1809), in the form of small spheres which for a part rest upon thick mycelium strands. This stroma is formed as in the former case [*Agaricus carneo-tomentosus*, the stroma of which, he says, at first quite homogeneous, is formed out of felted filamentous cells]. Gradually the same takes on an elongate form; the interior cells grow in a perpen-

[6] Handbuch der Allgemeine Mycologie 147–196. 1851.

[7] Pollinarien und Spermatien bei Agaricus. Bot. Zeit. **14**:137–148, 153–163. *pl.* 5. 1856.

dicular direction, the upper ones grow laterally, and then bend abruptly downward; the ends of these cells abjoint a parenchyma which forms the beginning of the lamellae; the under surface of these young lamellae is somewhat even and has no longer any connection with the stroma tissue lying close beneath, which later sinks down as a ring. The hymenial layer opens here round about at the side."

A few years later[8] he describes the development of a number of additional forms, all of which were gymnocarp except one, *Marasmius oreades*, and he remarks (p. 401) that this peculiarity of the internal origin of the hymenium is characteristic of many other Agaricaceae, as was shown in his larger work[9] which appeared in the following year.

The work of HOFFMANN was followed by DEBARY'S[10] study of the development of several species of Agaricaceae and Gastromycetes, by HARTIG'S study of *Agaricus melleus*,[11] by BREFELD'S studies on species of Coprinus,[12] and by the incomplete study of a large number of Agaricaceae by FAYOD[13] nearly twenty years ago, which was the last serious attempt (which I have as yet seen) at a study of the development of the Agaricaceae. Even these few studies do not agree in the account of development which they give of the same or related species. Furthermore, they are very incomplete and unsatisfactory, owing either to the methods employed (freehand sections by the earlier students), or in the scanty material at hand, which did not provide sufficient numbers of the early stages of development or a sufficiently full series of stages.

The difficulties in method are now overcome, but the other difficulties, those of obtaining sufficient material in all stages of development, are still serious in the great number of species. This is because

[8] HERMANN HOFFMANN, Beiträge zur Entwickelungsgeschichte und Anatomie der Agaricineen. Bot. Zeit. **18**:389–395, 397–404. *pls. 13–14.* 1860.

[9] Icones Analyticae Fungorum, Abbildungen und Beschreibungen von Pilzer mit besonderer Rücksicht auf Anatomie und Entwickelungsgeschichte 1–105. *pls. 1–24.* 1861.

[10] Morphologie und Physiologie der Pilze, Flechten und Myxomyceten 68. 1866.

[11] Wichtige Krankheiten der Waldbäume, etc. 25. 1874.

[12] Unters. a. d. Gesammtgebiet d. Mycol. **3**:13–122. *pls. 1–9.* 1877.

[13] Prodrome d'une histoire naturelle des Agaricinées. Ann. Sci. Nat. Bot. VII. **9**:181–411. *pls. 6–7.* 1889.

of the fact that at present few species have been cultivated artificially (except in the genus Coprinus by BREFELD, *l. c.*) so as to obtain the stages of development, and because the feral species nearly all pass their early and critical stages of development within the substratum and therefore are difficult to find, and at the same time it is often difficult to be certain to what species they appertain. These difficulties have probably played an important part in discouraging the further study of development of the Agaricaceae.

It is rather surprising, however, that even in the present time we do not have a sufficiently clear, full, and accurate account of the development of the fruit body of *Agaricus campestris*, especially the origin and differentiation of the various parts of the plant. This is the more so because this species is so common and of such wide distribution, but especially because it has been cultivated for so many years under conditions in which large numbers of carpophores in all conceivable stages of development are so easily obtained. Perhaps the very commonness and richness of the material has been the chief reason of its having been passed by.

Having given some attention to the study of the Agaricaceae for several years, especially as to their economic and biologic significance, as well as to the recognition of species and genera, the need of studies of development has been brought very forcibly to my attention, and I have been obtaining material for this study in several different genera. The meager and conflicting accounts which we have of the development of *Agaricus campestris*, as well as the ease with which material can be obtained, has led me to deal with this species first.

DEBARY[14] says the fruit body of many Agaricaceae (*Agaricus campestris*, *A. praecox*, *Coprinus micaceus* and relatives) is in its early youth a body interwoven out of delicate, dense, and uniform hyphae. At a very early stage, through differentiation of the original homogeneous weft, the principal parts of the fruit body are outlined and limited. On the interior of the upper part of the body a small and narrow air space of the form of a horizontal ring arises through the separation of the tissue elements. The portion which lies above becomes the pileus, the tissue present surrounded by it and below it

[14] Morphologie und Physiologie der Pilze, Flechten und Myxomyceten 68. 1866. Leipzig.

becomes the stem. The tissue on the outside of it corresponds to the edge of the pileus, but its hyphae continue without interruption or change into the outer surface of the stem below.

In 1874 R. HARTIG,[15] in his study of the development of *Agaricus melleus*, says that "the investigation of the earliest condition of the fruit body of *Agaricus melleus* shows that here, as well as in the Agaricaceae not provided with a veil, the pileus arises through a superficial annular furrow which in the beginning is completely open to the outside, and that later through growth of the marginal hyphae of the pileus and of the stem the annular furrow becomes covered over with a hyphal layer, the veil." He further says that if one compares his *fig. 20* (which shows the veil covering the hymenial tract) with DEBARY'S *fig. 26* (*l. c.*) of the young *Agaricus campestris*, "it appears from the agreement of the two figures that the conjecture is justified that also by this last fruit body in the region of the hymenial tract a subsequent growing together of the hyphae of the pileus and stem has taken place." Longitudinal sections of the young fruit body of *Agaricus campestris* at this stage do give the suggestion that the veil originates as HARTIG described for *Agaricus melleus*. DEBARY evidently did not study the very young stages of *Agaricus campestris*, for all his figures of longitudinal sections show the veil covering the hymenial area. It seems that without reinvestigating the question he adopted HARTIG'S suggestion that the development in *Agaricus campestris* followed the same course as that described by HARTIG for *Agaricus melleus*. In his later work[16] he says in reference to those forms with a "marginal veil" (*velum partiale* of FRIES): "Up to the first formation of the pileus on the summit of the stipe-primordium the phenomena are the same in essential points as in the gymnocarpous forms" (referring to his figures of *Collybia dryophila*, p. 55). "The young pileus is entirely delimited from the stipe by a transverse annular furrow running along its future hymenial surface. But then the superficial hyphal layers of the stipe and of the young pileus send out numerous branches toward one another from the edges of the furrow; these unite into

[15] Wichtige Krankheiten der Waldbäume, etc. 25. *pl. 106.* 1874.

[16] Comparative morphology and biology of the fungi, etc. 289–290. 1887. English edition.

a close weft, the marginal veil, which bridges over the furrow and closes it on the outside" (referring to figure of *Agaricus melleus* copied from HARTIG). To be certain that DEBARY here refers also to *Agaricus campestris* I quote also from page 291: "Most marginal veils are formed in the same way as that of *Agaricus melleus*, and *fig. 132* of *A. campestris* will serve to illustrate these remarks." *Fig. 132* is reproduced from his original work in 1866. In addition, on page 295 he says: "The account given above of the development of the species which are furnished with a marginal veil is founded, wherever it departs from my former statements, chiefly upon the facts discovered by HARTIG and BREFELD;" and on page 297, after discussing the different types of development in the Agaricaceae and his former statements, he says: "So far as these statements related to Coprinus they have been shown by BREFELD'S researches to be incorrect; my own did not pay sufficient regard to the earliest stages of development. I will not even maintain that they are quite correct for *Agaricus campestris*, ... but readily allow that the facts in the case are always the same as in *A. melleus*, and that the first extension of the marginal veil over the hymenial surface which was originally exposed had there also been overlooked."

GOEBEL[17] says: "These veil-formations are connected with the entire growth of the fructification; the species with a naked pileus are by their nature gymnocarpous." In speaking of the young fructifications of *Agaricus campestris*, he says: "These are at first pear-shaped bodies composed of young uniform hyphae, and each of these bodies is a rudimentary stipe, from the upper part of which the pileus will be developed. At an early period the hyphal tissue gives way in such a manner as to form an annular air cavity beneath the summit of the stalk, this cavity enlarges with the growth of the whole body, its upper wall forming the under side of the pileus, from which the radial hymenial lamellae grow in downward direction and fill up the cavity." His account thus supports DEBARY'S earlier account, but the evidence presented in illustrations is not sufficiently convincing in view of the controverted nature of the question and especially in view of the fact that gross anatomy and freehand sections

[17] Outlines of classification and special morphology of plants 132–134. 1887. English edition.

do not show the early stages of organization and differentiation, and GOEBEL's figures (*fig. 89, l. c.*) of the section of the young fruit bodies are made after the young pileus is differentiated from the stipe. No evidence is given that this text book account is the result of original investigation, and it is more likely that GOEBEL here is relying on the early account given by DEBARY, and the still earlier one given by HOFFMANN cited above.

In a similar way, in describing the early stages of development of *Agaricus campestris*,[18] I have followed DEBARY's later account as follows (p. 7): "At the same time a veil is formed over the gills by threads which grow from the stem upward to the side of the button, and from the side of the button down toward the stem to meet them. This covers up the gills at an early period." Aside from the extensive work of FAYOD in 1889,[19] little work seems to have been done on the development of the young sporocarps of the Agaricaceae since the publication of DEBARY's work in 1887.

In January 1905 an excellent opportunity presented itself for obtaining material in the required stages of development. Cultures had been made in boxes in the greenhouse of the commercial variety of *Agaricus campestris* known as Columbia, sold by the Pure Culture Spawn Company of Missouri. In many cases large numbers of young carpophores were formed at the surface of the substratum which were clean and in excellent condition for study. Preliminary examinations were made by freehand sections, and by staining in order to determine the age and size of the fruit bodies which should be prepared. In fruit bodies about 1^{mm} in diameter there was no evidence of a superficial annular furrow nor of any internal differentiation. Fruit bodies, therefore, from 1^{mm} to 4^{mm} were selected, fixed in chromo-acetic acid, imbedded in paraffin, microtomed, and stained, some in acid fuchsin and some in methyl blue.

The youngest stage is the primordium of the carpophore, a homogeneous body composed of slender uniform dense hyphae, intricately interwoven, and surrounded by a very thin layer of hyphae of a

[18] Studies in American fungi, mushrooms, edible, poisonous, etc., 1st edition, Ithaca, N. Y., 1900; 2d ed. 1901; and New York 1903.

[19] Prodrome d'une histoire naturelle des Agaricinées. Ann. Sci. Nat. Bot. VII. 9:181–411. *pls. 6–7.* 1889.

looser and less dense arrangement. This layer is the "universal veil." It is quite distinct in the young stages of these cultivated varieties, continues to increase in extent until the parts of the fruit body are differentiated and the young pileus and stem are manifest by external differences in form. Then it ceases to grow and is torn apart into white floccose patches on the pileus, as will be seen later. In the very young primordia then there is no evidence of a differentiation into stem and pileus in any of the many individuals which I have examined. As the primordia become slightly larger and older, but still before there is any evidence on the outside of an annular furrow or of any differentiation into pileus and stipe, longitudinal sections which are stained show two small deeply stained internal areas near the upper end of the young fruit body and some distance in from the surface. The hyphae here are not yet differentiated, but are richer in protoplasm, showing the origin of a new and special center of growth. This area is an annular one within the fruit body. Very soon afterward this area increases somewhat in extent and many hyphae begin to grow from the upper portion of this area downward. This is the primordial layer of the hymenium. It first arises when the tissue of the fruit body is homogeneous and compact except for the loose thin envelop. The hyphae which grow downward at this early stage are quite characteristic. They are slender and terete, tapering out into a long slender point. This enables them to pierce between the other hyphae of the compact fruit body. In fact at this time there are similar hyphae in the more central and upper portions, where the stem and pileus are to be differentiated, but in no other place at this time is there a definite center of growth which indicates the organization of any special part. These hyphae, partly at least, provide for intercalary growth of the young fruit body. Soon after the hyphae in the primordial layer of the hymenium begin to grow downward, there is a cessation of growth just below this area which results in the rupture and separation of the hyphae at this point in a corresponding internal annular area, forming the well known "gill cavity," which at first is very minute.

This annular primordium of the hymenium marks the differentiation of the primordium of the fruit body into the primordium of the pileus and that of the stem and veil, the latter being the tissue of

the young fruit body external to the hymenial primordium and continuous with what is to be the margin of the pileus above and with the undifferentiated stem surface below. The downward growing hyphae now take on a different form. They are still slender, but are even and blunt and are very densely crowded side by side, are very rich in protoplasm compared with the hyphae of the rest of the young carpophore and consequently take a deep stain.

This first growth takes place on the under side of the young pileus primordium. The vegetative activity in this region of the young pileus is very great, and is very soon extended outward on the periphery or margin of the young pileus, as shown by the very rapid radial growth of the hyphae at the margin of the pileus, but still some distance in from the surface. This radial growth is also accompanied by a very strong hyponastic growth, so that the threads curve downward, and soon it is so strong that the margin of the pileus is strongly incurved, the hyponastic growth appearing to be stronger at the margin and near it than further inward.

At the same time the primordium of the marginal veil increases by intercalary growth. In the participation of the hyphae at the margin of the pileus in the formation of the veil, they seem to show a greater activity in growth so far as the density of the growth and richness in protoplasm is concerned; while the larger portion toward the stem, also increasing by intercalary growth, becomes looser by the rapid elongation of its elements and their partial separation, thus forming numerous small air spaces. This seems to have an important bearing on the supply of fresh air to the young forming hymenium where the air spaces become continually larger, and the first air space formed underneath the annular primordium of the hymenium not only gives place for the development of the latter but also provides aeration. Thus while the veil serves the purpose of protection to the young hymenium, its structure is such as to provide aeration also. After the differentiation of the hymenial primordium, the lateral growth of the pileus is accentuated so that it becomes broader than the stem portion, and now is formed the external annular furrow. Very soon after the hymenial primordium is organized, the tissue of the pileus primordium, or end of the fruit body, takes on a deeper stain in an area extending inward and some depth below

the surface, showing that the pileus primordium is becoming definitely organized through the central portion (*fig. 4*), also sometimes seen in freehand sections in older stages as a compact area (*fig. 15*).

In the very early stage of the hymenial primordium the under surface is even, but very soon the outlines of the gills begin to form by the more rapid downward growth in radial lines. Very soon after the young gills begin to form, the surface of the stem is differentiated. This occurs in such a way as to show that the surface of the stem portion of the young fruit body does not become the surface of the stem except at its extreme lower portion, which probably corresponds to the bulb even when a bulb is not manifest as a thicker portion of the stem. From a point at the junction of the original annular primordium of the hymenium, or the junction of the young stem with the pileus at the inner end of the gills, and then extending obliquely downward and outward, the hyphae take on more active intercalary growth and a richer content of protoplasm. This marks the origin of the cortex of the stem, and distinguishes it very sharply from the elements of the veil and from the internal tissue of the stem which is to be the medulla by a deeper color in stained preparations; also in fresh freehand sections it is usually very clearly seen with the unaided eye as a whitish more compact area which shows well in ordinary photographs at this stage.

This study shows very clearly that the hymenium in *Agaricus campestris* is endogeneous in its origin, as HOFFMANN described so early as 1856 (*l. c.*), and that DEBARY'S first account of the development of this species in 1866 was correct so far as it went; but he did not succeed in obtaining for study carpophores sufficiently young to enable him to speak with certainty after HARTIG had thrown doubt on his conclusions as a result of his study of *Agaricus melleus*.

Not only is the hymenium endogenous in its origin, but the differentiation of the stipe and pileus is simultaneous; that is, the initial stages of the stipe and pileus as distinct structures are organized and made evident in longitudinal sections by the origin of the hymenial primordium. In all the sections that I have examined of this species at this stage, there is no evidence of differentiation of the pileus from the stem before the earliest evidence of the hymenial primordium. We should not conclude, however, this mode of development

is necessarily to be found in other plants until they have been studied, though it is probable that it is true for at least some of the other species of Agaricus (Psalliota). FAYOD found that usually the primordium of the pileus was organized in the apex of the homogeneous young carpophore before the appearance of the primordium of the hymenium, and this seems to be true in certain species of Hypholoma studied by one of my students. In fact FAYOD asserts that this can be accepted as a general law. The primordium of the pileus is the first to appear in the organization of the parts of the carpophore from its primordium. It is shown by the area of hyphae composing it taking on a deeper stain in sections. It is in the form of an inverted bowl convex above, concave below, and *Agaricus rubellus* (*Psalliota rubella*) is one of the forms which he studied. The only exceptions which he admits are the coriaceous forms like Lentinus (*l. c.*, p. 296). In this respect these cultivated forms of *Agaricus campestris* show also an exception to this rule, and the primordium of the pileus is to be regarded as diffuse in the primordium of the carpophore, as he suggests for the coriaceous forms.

The question of the simultaneous organization of the pileus and stipe from a young homogeneous fruit body is an interesting one in view of the different theories held by some of the earlier students. Thus FRIES[20] said: "Omnia organa simul, nulla subevolutione nova successive explicantur. Omnes extremitates ipsius Fungi explicati jam in aetate juvenili adsunt," *i. e.*, "all organs are unfolded simultaneously, none by new successive development. All parts (or extremities) of the unfolded fungus itself are already present in the young stage." He thus believed that in the very young fruit body the organs or parts, though rudimentary and invisible, were all present, their manifestation and expression was a matter of unfolding.

This interesting conception is shown also in another place (*l. c.* 40), where he expresses his view of the general mode of growth of the fungi as compared with that of the algae as follows: "Fungi in omnes plerumque directiones a centro, quod junius, sese expandunt," *i. e.*, "Fungi, though young, expand from the center in almost all directions." All the parts being present, the growth in the center pushed them outward, as they enlarged, into their respective positions, while

[20] E. FRIES, Systema orbis vegetabilis, part 1, 40, 43.

the growth in the case of the algae was at the periphery—"explicantur ita, ut extremitates semper sint juniores" (*l. c.* 53).

Then as to the relative time, or priority of the shaping of the different parts, FRIES says (*l. c.* 44): "Pileus v. c. in Agarico formatus est prius quam stipes pronascitur. Stipes enim pilei, cum quo contiguus, prolongatio et receptaculi pars," *i. e.*, "the pileus, for example, in Agaricus is formed before the stipe is produced. The stipe certainly is a prolongation of the pileus with which it is contiguous, and part of the receptacle."

J. SCHMITZ[21] in his *Mycologische Beobachtungen*, as a result of the study of several forms, holds an entirely different view. He agrees that according to his own observations in many cases all parts of the fungus are formed and present in a very young stage. But he cannot understand, nor believe, that these parts are formed at once, that is, simultaneously with the origin of the fruit body as if by the touch of a fairy wand ("wie durch einen Zauberschlag," p. 174). That at certain young stages all parts are to be considered present and yet invisible he regards as belonging to the domain of pure speculation, a hypothesis suited to a philosophical mode of representation or idealization, and not corresponding to reality. SCHMITZ gives an account of his studies on *Cantharellus sinuosus*, *C. tubaeformis*, *Agaricus bulliardi* Pers. (=*Cortinarius bulliardi* [Pers.] Fr.), *Coprinus niveus*, and *Hydnum repandum*, and believes he is justified in formulating for the pileate fungi a law of development as follows: "1st, that a successive formation of single parts or organs takes place; 2d, that this appearance of new parts rises upward just as gradually as in the case of the higher plants, in such a way that the higher standing parts naturally come to view later than the lower parts, that the matrix or hypothallus appears before the stipe, the stipe before the pileus, and the pileus before the hymenium.[22] In *Agaricus* (*Crepidotus*) *variabilis*

[21] Ueber die Bildung neuer Theile bei den Hymenomyceten, vorzugsweise den Pileaten. Linnaea **16**:168–179. 1842.

[22] "Ich glaube also, dass man bei der Pileaten als Gesetz aufstellen darf, dass 1) eine successive Bildung der einzelen Theile oder Organe vor sich gehe; 2) dass dieses Hervortreten der neuen Theile ebenso graduell aufwärts steige, wie bei den höhern Pflanzen, so nämlich, dass die höherstehenden wesentlichen später als die untern zum Vorschein kommen, also die Matrix vor dem Stipes, dieser vor dem Pileus und der letztere (an und für sich) vor dem Hymenium auftrete."

OERSTED[23] says that the stem is produced first, and afterward the pileus.

In the forms of *Agaricus campestris* studied here, as I have shown, the young homogeneous fruit body (*figs. 1, 2*) shows no differentiation into parts (except the rudimentary universal veil), and it is to be considered as the primordium of the carpophore. It is not a stem, nor is it a pileus, there is no differentiation to show even the rudiments or primordia of stem or pileus; there is no more active growth manifest in one place than in another, and no separate group of hyphae with richer protoplasmic content which gives a differential stain. It cannot therefore be considered as a rudimentary stem, as GOEBEL (*l. c.* 132–138) has suggested. It is true we might speak of a stem end and a pileus end, but the application of these terms to the portions of the carpophore primordium which are later to be organized into pileus and stem primordia does not predicate their existence before organization takes place. But soon the differentiation takes place by the appearance of the primordium of the hymenium, which at once delimits also the primordia of pileus, stipe, and marginal veil (*figs. 3–5*). This condition answers well to the conception of FRIES that all parts are present in the young stage, though he conceived them to be present in the still younger stages, which we find is not the case.

DECANDOLLE[24] says that in the case of Agaricus the upper part or pileus develops before the lower part, or stipe. Without critical study of the very young stages one might be led to this conclusion by an examination of certain large pileate short-stemmed forms, and perhaps DECANDOLLE examined such plants. According to FAYOD (*l. c.* 279–280) the pileus is differentiated first, the hymenium

[23] OERSTED, A. S.: Iagttagelser anstillede i Löbet af Vinteren 1863–64, som have ledet til Opdagelsen af de hidtil ukjendte Befrugtningsorganer hos Blapsvampene.—Oversigt over det Kongelige danske Videnskabernes Selskabs Forhandlinger, p. 11. *pls. 1–2.* 1865. Copenhagen.

See translation, Observations made in the course of the winter of 1863–64, which have led to the discovery of the hitherto unknown organs of fructification in the Agaricini by A. S. OERSTED. Quart. Jour. Microscop. Sci. **8**:18–26. 1868.

[24] "Dans plusieurs, tels que les Agarics, la partie supérieure, qu'on nomme le chapeau, paraît développée avant l'inférieure, qu'on a comparée à une tige ou à un pédoncle; l'inverse semble avoir lieu dans les Clavaires qui paraissent croître de bas en haut." Organographie végétale **1**:384. 1827.

and stipe later, and it is interesting to note that W. G. SMITH[25] in his study of *Coprinus radiatus* says that the cells of the pileus and the hairs which form the veil are the first to appear (*l. c.* 62), but his study does not seem to have been exact, and a comparison of such a form as Coprinus with Agaricus (Psalliota) is not pertinent at present except as it bears on the attempt of some to formulate a general law of growth.

If we now turn to the law formulated by SCHMITZ for the order of succession of the different parts of the pileate fungi, we see that *Agaricus campestris* does not conform to it, but that it is more in accordance with the idea expressed in the first sentence quoted from FRIES, that all parts of the fungus unfold simultaneously. This must not, however, be taken wholly to support FRIES' conception of the young sporocarp nor his idea of central growth. While it is likely that a number of other fungi will be found to agree with *Agaricus campestris* in the mode of organization of the parts of the plant from the primordium of the sporocarp, it is certain that no law of organization and succession of the parts can be formulated which will hold good of all the pileate fungi. There are probably some, as suggested above, in which the pileus and stipe primordia are organized before the primordium of the hymenium, and many others probably in which the stem is partially or quite well organized before there is even a primordium of the pileus, in which case the development would be in conformity with SCHMITZ's law given above. This is very likely the case with certain long, slender-stemmed species of Marasmius, of such plants as *Polyporus lucidus* Leys. (*Ganoderma lucida*), and others. But we must wait until the different types have been carefully studied from the very young stages in microtome sections.

FAYOD (*l. c.*), who studied a large number of Agaricaceae, formulates the general law that the pileus is organized first within the young primordium as a pileogenous layer (*couche piléogène*), which arises by internal differentiation, marked by the more rapid growth of the hyphal elements and their richer content in protoplasm. This layer is in the form of a shallow inverted bowl, convex above, concave below. This is surrounded on the sides and above by a thin layer

[25] Reproduction in *Coprinus radiatus*. Grevillea 4:53–65. *pls.* 54–61. 1875.

which he calls the primordial cuticle (*cuticle primordiale*). From his description the primordial cuticle varies in character and probably in structure also, and it is difficult to accept his conception of a primordial cuticle as a homologous structure in the large series of forms to which he applies the term. For example, he recognizes three main types in the development of the Agaricaceae: 1st, the gymnocarpous forms; 2d, the angiocarpous forms; 3d, the endocarpous forms, and the primordial cuticle is present in all except in a very few of the gymnocarpous forms. In the gymnocarpous forms the primordial cuticle consists generally of a more dense layer of tissue underneath which the pileogenous layer is formed. The margin of this becomes the border of the pileus, and as it extends laterally it dislocates at this point the primordial cuticle, so that the primordium of the hymenium which now arises is of exogenous origin, thus giving rise to the gymnocarpous type. Examples are *Panus stipticus*, *Cantharellus cibarius*, *Marasmius rotula*, *Collybia racemosa*, etc. Thus while DEBARY believed the pileus in the gymnocarpous forms originated exogenously, FAYOD holds that it originates endogenously; but in discussing farther certain other forms he admits that the pileus is formed in the manner indicated by DEBARY, and he states that the discussion which he has raised here is more in regard to a principle than fact, and he would not have raised the question at all had it not been for the fact that his study of the angiocarpous forms had shown him the importance of the pileogenous layer.

In many of the angiocarpous forms the primordial cuticle would seem from his own description to be a different structure from what it is in the gymnocarpous forms, for he says: "The hyphae which emanate from the pileogenous layer do not reach the surface of the primordium. As a consequence the primordial cuticle, which acquires here a very considerable thickness, preserves its integrity and continues to increase up to the time of the formation of the lamellae and stipe, that is, up to the second period of development." Here he recognizes the primordial cuticle as identical with the universal veil, and he would call it general veil (*voile général*), although it is often formed in some cases by parts also of the subjacent tissue of the primordium, were it not for the fact that he wishes to place it in the same category as the non-integral element of the primordium

in the gymnocarpous forms which he considers a primordial cuticle.

The angiocarpous forms he further divides into two types, subangiocarpous and angiocarpous. In the subangiocarpous type the universal veil (*cuticule primordiale*), being continuous over the pileus and stem, forms the veil which is known as the "partial veil" or "marginal veil" of authors. He says (p. 286) it is probably characteristic of Flammula, Inocybe, Dermocybe, Hygrocybe, Psalliota, Lepiota, Psathyrella, Coprinus, and most of the Tricholomae. In the true angiocarpous types there is a cuticle of the pileus which is organized underneath the primordial cuticle or universal veil, so that at maturity the universal veil separates and forms floccose patches, or a volva, or may disappear by gelatinization. As examples he cites Agrocybe (*Pholiota praecox*, *Naucoria semiorbicularis*, etc.), Pholiotina (*Pholiota blattaria*, *P. togularis*, etc.), *Rozites* Karsten (*Pholiota caperata*), *Nemataloma* Karsten, some species of Panaeolus, Telamonia, and probably Locellina and Chitonia; the volva in the last three genera he considers to be only a very thick universal veil.

In the endocarpous forms the primordium of the fruit body is differentiated on the interior of a primitive bulb which he calls the primordial bulb (*bulbe primordial*), to which belong the greater number of species of Amanita, Volvaria, and some species of Phlegmacium. Since this type does not concern us here it will not be in place to discuss it.

From the foregoing it is seen that FAYOD places Psalliota (which includes *Agaricus campestris*) in his type of subangiocarpous forms. Among these forms he studied *Agaricus rubellus* Gillet (*Psalliota rubella*). While he does not describe the development of this species (his discussions of development are in the form of general conclusions), he says that it belongs to the subangiocarpous type, and his *fig. 4, pl. 7*, shows the primordial cuticle to consist of rather loose radiating threads connected on the sides where it extends down over the lamellae and stipe with the thicker portion covering the stem. Although FAYOD placed *Agaricus campestris* also in his subangiocarpous type, a study of these cultivated forms shows that it would belong to his true angiocarpous type because of the free universal veil entirely independent of the marginal veil, the universal veil eventually sepa-

rating into floccose patches on the surface of the pileus as in *Rozites caperata* (Pers.) Karsten (*Pholiota caperata* Pers.).

During the later period of growth and the beginning of elongation of the plant, the marginal veil increases in thickness and extent. It is entirely free from the lamellae, the hymenial cavity being quite distinct from the first and becoming greater by the expansion of the pileus and marginal veil, and also by the elongation of the portion of the stem above its attachment. The increase in the surface extent of the marginal veil is considerable and results in throwing the upper surface into radiate folds which are quite noticeable, especially in the well developed individuals. In the young primordium at the time of the organization of the parts of the carpophore the marginal veil is attached over a large part of the outer surface of the stem primordium, the lower end, perhaps that portion which corresponds to the bulb in other species, being free. It thus remains attached over the stem surface for a considerable period during growth. As the period of elongation advances, the veil begins to separate from the stem at the lower end and is gradually torn off and upward as the pileus expands and the stem elongates. The tension of the connecting fibers can very easily be seen between the stem and the under surface of the veil, and is well shown in *fig. 20*. It therefore forms a sheath over the stem except a short section of the lower end, and the portion above the marginal veil which is elongating. This sheath is loosened from below upward except at the upper point at attachment to the stem. The outer margin of the veil is attached to the rounded and thick margin of the pileus, and being of considerable thickness in these cultivated forms the lower edge of the veil is separated first from the outer surface of the pileus margin (*fig. 40*), and the inner upper edge is separated last from the inner surface of the pileus margin. The margin of the veil is therefore furrowed (*figs. 18, 19*). A thick marginal veil of this type is called a "double veil," a type which is very characteristic of certain other species of Agaricus where it is more highly developed, especially in *Agaricus arvensis*, where the lower portion of the veil splits radially. It is very striking in *Agaricus rodmani* Pk., where the forking of the veil extends almost to the stem.[26] In *Agaricus placomyces* Pk. the veil is often similar to

[26] See *fig. 17*, ATKINSON, Studies of American fungi, mushrooms, edible, poisonous, etc. 1900, 1901, Ithaca; 1903, New York.

that of *Agaricus arvensis* (*figs. 21, 22* in Studies Am. fungi, etc.), while in *Agaricus silvicola* and others the lower half of the marginal veil is often separated into patches (*l. c. fig. 20*). In the pasture or field forms of *Agaricus campestris* the marginal veil is thinner, but even here its double character is often manifest (*fig. 7, l. c.*).

The growth of the pileus which at first is strongly hyponastic becomes less so as the pileus expands. The upper surface gradually ceases to grow and the extension of the underlying part often tears the pileus cuticle into fibrous scales. The growth of the pileus gradually becomes epinastic, as the lower area and the hymenophore with the gills now grow more rapidly than the middle and upper portions. This causes the pileus to become plane, or in old specimens the margin itself becomes upturned. This peculiarity in the growth of the Agaricaceae during the period of elongation was supposed by some of the earlier botanists[27] to be due to the influence of light, for it was thought by them to be necessary that the hymenium should be turned up to the light. We now know that light is not necessary for the growth and ripening of many species. This partial eversion of the pileus in many species unquestionably serves a useful purpose in providing for the wider distribution of the spores, for they are more easily caught by currents of air as they leave the hymenium.

The order in which elongation of the different parts takes place is thus different from the order of their initiation in the young primordium. As has been shown in these cultivated forms of *Agaricus campestris*, the organization of the primordia of pileus, stem, hymenium, and marginal veil is practically simultaneous by the appearance of the hymenial primordium as an internal annular area; while the organization of the parts gradually proceeds and is also simultaneous to a certain degree. But the period of elongation of the parts after they have become organized, while overlapping to a certain extent, follows in succession. The marginal veil completes its period of elongation first, then the stem, followed by the pileus, and finally the hymenium.

One striking feature of the hymenium of these cultivated forms is that, so far as I have examined (the varieties Columbia, Alaska,

[27] See NEES VON ESENBECK, Das System der Pilze und Schwämme 179–187. 1816.

Bohemia, and others), the basidia are two-spored. I have several times observed this fact in the cultivated mushroom. The illustration of the hymenium of *Agaricus campestris* which I have used on two former occasions[28] was made from a cultivated variety. GOEBEL'S *fig. 90*[29] shows only two spores on the basidium of *Agaricus campestris*, and this was probably also made from a cultivated variety. I have on the other hand several times observed that in case of the normal field or pasture form of *Agaricus campestris* there are four spores on a basidium. The nuclear phenomena in the formation of the spores have not yet been thoroughly worked out in the two-spored forms of *Agaricus campestris*, but studies of C. E. LEWIS carried on in my laboratory seem to show that the normal number of four nuclei are first formed in the basidium, and that two of them degenerate. This has been very clearly shown by him to be the case in a new species of Amanita, *A. bisporigera* Atkinson.[30] Nor has it been shown how the two-spored forms of *Agaricus campestris* arise from the normal four-spored feral plant, or whether normal two-spored forms exist as constant types in the field under natural conditions of environment. I have found a two-spored Agaricus resembling in some respects certain of the cultivated forms of *Agaricus campestris* growing spontaneously in the open. On one occasion it was found in June about young trees in a lawn which had been mulched with horse manure. On another occasion the same species was found on the hillside of a wooded ravine (Cascadilla gorge) on the campus of Cornell University.

If there are two-spored forms of *Agaricus campestris* existing under natural conditions of environment which are constant and which present also other characters even slightly different, it would indicate that *Agaricus campestris* either is or recently has been passing through a mutating period, and that these forms are elementary species. Were the two-spored basidia the only differentiating character, such a form might in the sense of DEVRIES[31] be regarded as

[28] Studies and illustrations of mushrooms. I. Cornell Univ. Agr. Exp. Sta. Bull. 168. *fig. 189.* 1897; and Studies of Am. fungi, etc. (*l.*)

[29] Outlines of classification and special morphology, Eng. ed., 1887.

[30] LEWIS, C. E., The development of the spores in *Amanita bisporigera* Atkinson. BOT. GAZ. 41:348–352. 1906.

[31] Species and varieties, their origin by mutation. 1905.

a variety, for it would seem that the four-spored quality or character is latent, since four nuclei are probably formed in the basidium in the normal manner but only two of them function. With regard to the cultivated forms of *Agaricus campestris* they probably represent also mutations either from *Agaricus campestris* or from some other species which has been confounded with it. Whether they are to be considered elementary species or varieties or retrograde varieties would depend upon their constancy or inconstancy, their stability or instability. They may be horticultural or domesticated varieties. Nevertheless it would seem that they have arisen by mutation. It is interesting to note in this connection that, whether species or varieties, if they have arisen by mutation their chances of becoming constant may be greater than in the case of plants which are well known to reproduce sexually. It is generally believed that the Agaricaceae are not reproduced by the cooperation of sexual organs. If this is true, and if there is no process similar to fertilization, mutations of these plants would escape one of the operations in nature against the constancy of new mutants in species capable of cross fertilization. Some students regard the fusion of the two primary nuclei in the basidium as an act of fertilization, but from what we know of the origin of these two nuclei the possibility of cross fertilization of individuals at this epoch of development is excluded, though it cannot at present be regarded as impossible at an earlier stage in their ontogeny. Of course the earlier ideas of fertilization in the Agaricaceae held in the time of BULLIARD,[32] who called the cystidia spermatic vessels and thought they squirted their juices on the seeds (spores) thus bringing about fertilization, or by CORDA[33] who regarded the cystidia as pollinaria and thought fertilization was brought about by the exudation of their fluid content to which the spores became attached and fertilized, are now unthinkable, as well as the notion of W. G. SMITH[34] as late as 1875, who believed that filaments growing out from the cystidia came in contact with the spores and fertilized

[32] BULLIARD, Histoire des champignons de la France 1:39–66. 1791.

[33] CORDA, Berich. Ises. 6:40. 1834; also Icones 3:44. 1839. See also H. HOFFMANN, Pollinarien und Spermatien bei Agaricus. Bot. Zeit. 14:137–148, 153–163. *pl. 15.* 1856.

[34] SMITH, W. G., Reproduction in *Coprinus radiatus.* Grevillea 4:53–65. *pls. 54–61.* 1875.

them, and that hybrids between species were very commonly found where cystidia and spores from adjacent species fell to the ground, commingled, and brought about cross fertilization. But the last word may not yet have been said with reference to the possibility of a fertilization prior to or during the early stages of the organization of the primordium of the carpophore, like that proposed by OERSTED (*l. c.*) for *Agaricus* (*Crepidotus*) *variabilis*, or in some closely related manner.

However, the propagation of forms by spawn which is not obtained from the spores, as is practiced by DUGGAR,[35] would seem to be equivalent to vegetative propagation or budding, and thus might be of advantage in maintaining constancy in varieties, since they would not be subject to cross fertilization, though it is still a question if fertilization and cross fertilization take place in the Agaricaceae. If it does not, or if some process equivalent to it, especially cross fertilization, does not take place, the Agaricaceae, and in fact the Hymeniales, would be especially free from the production of hybrids, and the constancy of species or varieties arising by mutation would be correspondingly favored. In a number of species there are indications that mutation is now going on, or that these species have recently passed through a period of mutation, and some of these apparent mutants appear to be quite constant. On the other hand, there are many species which show great fluctuating variability due to varying conditions of food supply, moisture, substratum, etc.

CORNELL UNIVERSITY,
Ithaca, N. Y.

DESCRIPTION OF PLATES VII–XII.

Photomicrographs with Zeiss microscope except *fig. 11;* plate holder 360^{mm} from object on slide; photomicrographs and photographs by the author.

PLATE VII.

FIG. 1. Young carpophore, var. Alaska, undifferentiated; oc. 2, obj. 16^{mm}.

FIG. 2. Same as *fig. 1*, but with oc. 4, obj. 16^{mm}.

FIG. 3. Young carpophore, var. Columbia, with primordium of hymenium in earliest stages of endogenous origin; oc. 2, obj. 16^{mm}.

[35] DUGGAR, B. M., The principles of mushroom growing and mushroom spawn-making. U. S. Dept. of Agr., Bureau of Plant Industry, Bull. 85. pp. 9–60. *pls. 1–7.* 1905.

FIG. 4. Young carpophore, var. Alaska, with endogenous primordium of hymenium a little more advanced than in *fig. 3;* universal veil distinct as a loose definite layer of tissue surrounding the carpophore; oc. 2, obj. 16mm.

FIG. 5. Young carpophore, var. Columbia, showing endogenous primordium of hymenium a little more advanced than in *fig. 4;* universal veil as a very thin layer; oc. 2; obj. 16mm.

PLATE VIII.

FIG. 6. From young carpophore, var. Columbia; at stage when gill slit is just forming, showing sharp-pointed threads of primordium of hymenium projecting downward; also shows at the right the thin layer of universal veil; about same magnification as *fig. 7*, but not with Zeiss microscope.

FIG. 7. Same object as *fig. 6*, but showing only the gill slit and primordium of hymenium.

FIG. 8. Same as *figs. 6, 7*, but more highly magnified; oc. 6, obj. 3mm.

PLATE IX.

FIG. 9. Young carpophore, var. Alaska, showing endogenous primordium of hymenium on one side, about the same stage as *fig. 4*, but higher magnification; oc. 4, obj. 16mm.

FIG. 10. Young carpophore, var. Alaska, with endogenous primordium more advanced, showing definite and clear opening, the descending threads of the primordium of the lamellae, loose tissue of the inner veil, and primordium of stem cortex, outlined as an oblique area of younger threads rich in protoplasm, extending from opening obliquely downward and outward; oc. 6, obj. 3mm.

FIG. 11. Portion of young carpophore, var. Columbia, showing gill slit; young lamellae in longitudinal section, inner veil of more open loose tissue; shows also how primordium of hymenium continues in its development as margin of pileus continues to grow; cortex of stem well-formed, showing as a more compact tissue over the surface to which the veil is attached; oc. 4, obj. 16mm.

FIG. 12. Portion of young carpophore same age as *fig. 11* and from same plant, but cut somewhat obliquely so that a number of gills are shown in oblique section, otherwise as in *fig. 11*, but with less magnification; oc. 2, obj. 16mm.

PLATE X.

FIG 13. Cluster of young carpophore, var. Columbia; numerous very young ones, several in the large button stage, showing the small white patches of the universal veil on the brown cuticle of the pileus.

FIG. 14. Another cluster of young carpophores, var. Columbia, showing numerous very small ones, and several of the small button stage, the universal veil separated into a few large white thin patches; rhizomorphs in the substratum.

PLATE XI.

FIG. 15. Longitudinal section of young carpophores magnified twice the real length, showing endogenous primordium of hymenium, gill slits, veil, and the primordium of cortex of stems.

FIG. 16. Real size; young carpophores, var. Columbia, showing rhizomorphs, the expanding young pileus and universal veil separated into patches; it is very distinct in the two plants where the universal veil is stretched between the two caps; at the right a few in longitudinal section.

FIG. 17. Young carpophore, var. Columbia, about half grown, real size, showing a few white patches of universal veil on the pileus, about midway of the short stem showing a ring which is the lower part of the double ring; in this case separated from the upper part and remaining on the stem as a distinct ring as in *Agaricus rodmani;* upper portion of the veil still attached from stem to margin of pileus which is as yet close against the stem.

PLATE XII.

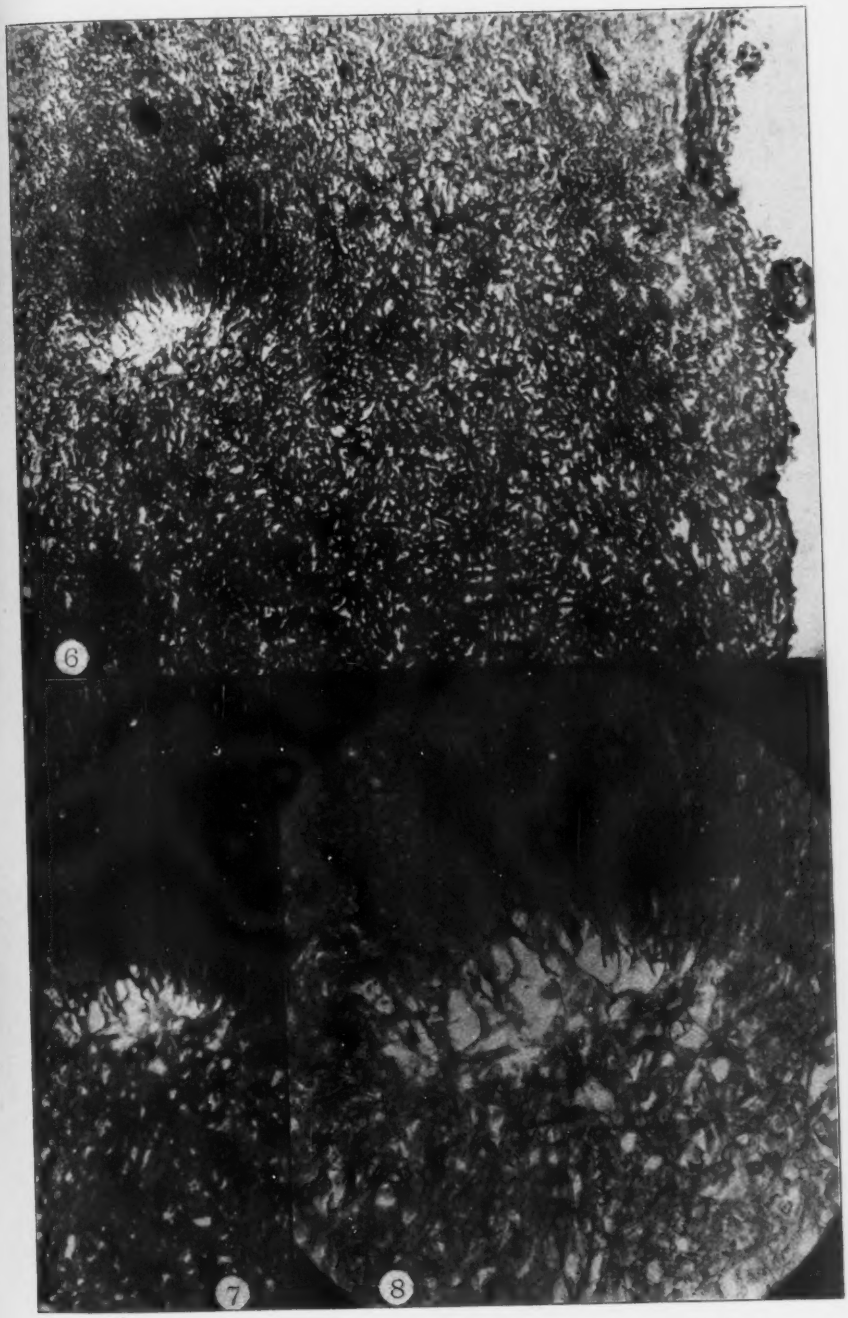
FIG. 18. Cluster of mature carpophores of *Agaricus campestris*, cult. var., showing patches of universal veil on pileus.

FIG. 19. Mature and nearly mature plants, var. Columbia, showing thick veil which forms a sterile margin on the edge of the pileus and a thick ring wrinkled or corrugated on the upper surface and the edge distinctly double.

FIG. 20. Slightly younger stage, also lower part of double veil as broken away from the outer surface of margin of pileus; upper part of double veil still attached to margin of pileus.

The material was selected and fixed in chromo-acetic acid by myself, and I am under many obligations to Dr. CHARLES E. LEWIS, who then took the material, carried it into paraffin, sectioned, and stained it.

ATKINSON on AGARICUS



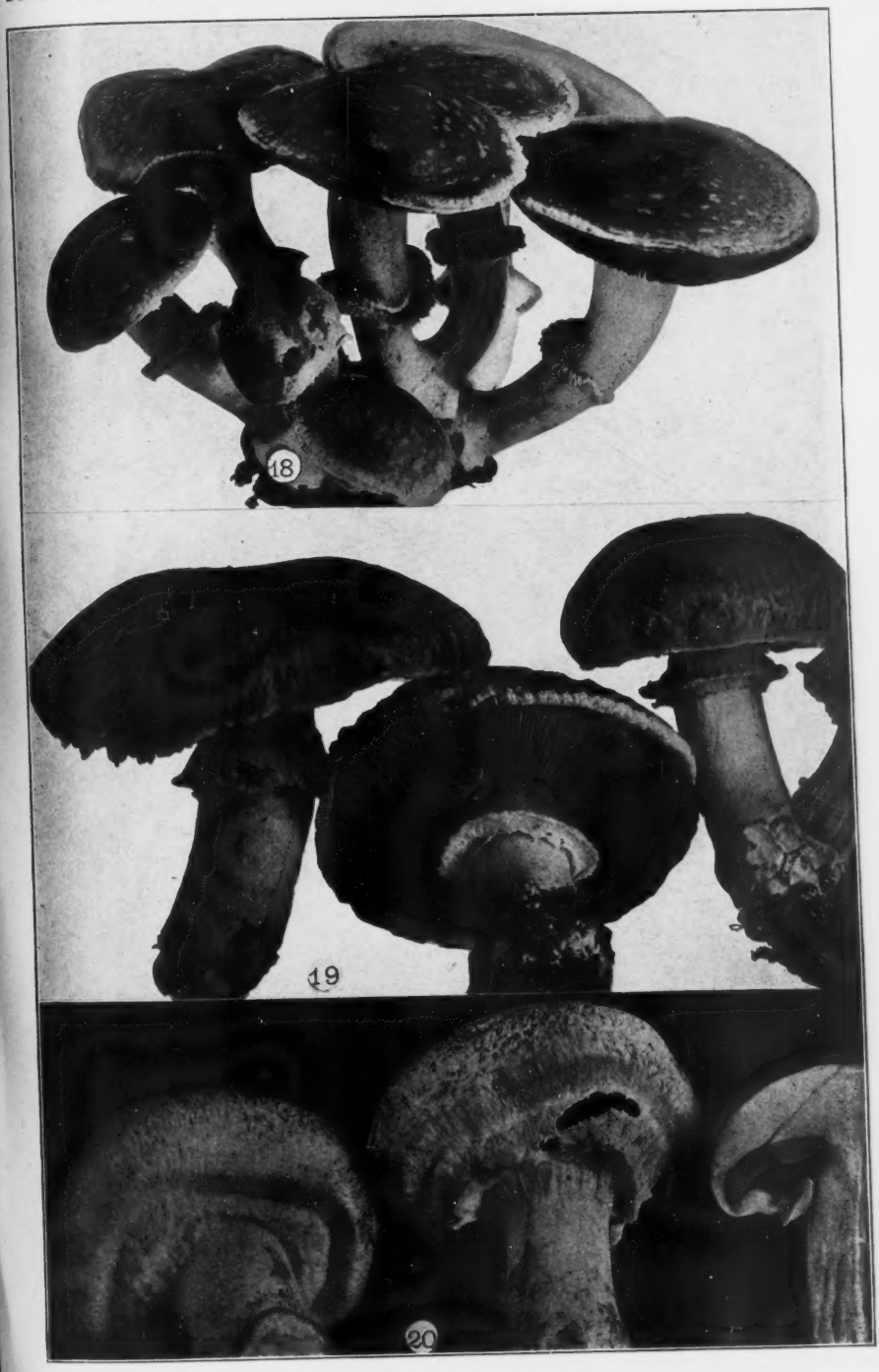
ATKINSON on AGARICUS

ATKINSON on AGARICUS

ATKINSON on AGARICUS

PLATE XI

ATKINSON on AGARICUS



ATKINSON on AGARICUS

RÔLE OF SEED COATS IN DELAYED GERMINATION.

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY. LXXXV.

WILLIAM CROCKER.

(WITH FOUR FIGURES)

I. Historical.

IT is well known that in many species of plants not all the seeds of a given crop germinate promptly after being subjected to so-called germination conditions; instead they germinate at irregular intervals through a period of weeks, months, or even years. It happens in many species that none of the seeds of a crop will germinate until they have been subjected to germinative conditions for a year or more, and that in these cases of marked delay germination is distributed through a further period of greater or less length.

Delayed germination is well illustrated in the results of the researches of NOBBE and HÄNLEIN (**8**, *a*, *b*). Table I shows their observations on thirty-one species of common weeds. They began with 400 seeds of each species and continued their experiments 1,173 days.

KIENITZ (**4**) found marked distribution in the germination of crops of the beech, white fir, ash, hornbean, and pine; and WINKLER (**15**) in sowings of *Euphorbia cyparissias*, *E. exigua*, Cuscuta, etc. WIESNER (**14**) found that the seeds of *Viscum album* germinate only sparingly in the fall after ripening, but readily the following spring. KUNTZE (**6**) in reviewing the literature on germination mentions a large number of cases of delayed germination. The hawthorn, he states, will grow only after being in the ground one to three years.

One of the most interesting cases of delayed germination is that of the cocklebur (Xanthium) reported by ARTHUR (**1**). He found that the two seeds in the bur are not the exact counterparts of each other, but can be distinguished readily by their form and position in the bur. One seed, which he terms the upper because it is borne nearer the apical end of the bur, is convex on the outer face and con-

TABLE I.

AFTER NOBBE AND HÄNLEIN; 400 SEEDS OF EACH SPECIES.

Name/No. of seeds germinated on day	4	5	6	7	8	9	10	16	36	72	145	351	519	714	874	1082	1173	Total	Per ct.
1 Aquilegia vulgaris	..	..	..	...	...	...	..	3	..	..	..	..	...	...	...	...	...	3	0.75
2 Campanula rotundifolia	..	..	..	11	8	1	1	1	1	..	..	..	11	13	10	1	...	58	14.50
3 Campanula persicifolia	..	..	..	...	3	8	14	112	56	1	..	11	9	2	8	2	6	232	58.00
4 Campanula Trachelium	..	..	..	...	...	...	..	...	..	..	..	..	1	...	...	...	...	1	0.25
5 Chaerophyllum temulum	..	..	..	...	...	...	..	...	..	..	..	..	...	...	...	...	3	3	0.75
6 Chenopodium album	..	..	2	...	...	2	3	2	..	1	19	53	27	...	2	3	3	117	29.25
7 Capsella Bursa-pastoris	..	3	..	3	...	...	..	...	..	..	4	4	20	...	24	10	7	75	18.75
8 Chelidonium majus	..	..	..	...	...	...	..	...	59	8	2	..	...	3	90	1	32	195	48.75
9 Digitalis purpurea	..	..	..	9	45	102	47	159	24	1	..	..	...	...	rest	eca	yed	387	96.75
10 Hypericum hirsutum	..	..	..	2	1	4	13	132	58	..	..	2	...	...	...	...	...	212	53.00
11 Hypericum montanum	..	..	..	3	19	61	31	171	46	..	..	..	...	1	...	...	...	332	83.00
12 Hypericum perforatum	..	..	..	1	...	...	2	20	28	2	..	..	...	1	3	1	...	58	14.50
13 Jasione montana	..	..	17	98	141	81	30	28	2	..	..	..	...	...	rest	deca	yed	397	99.25
14 Lithospermum arvense	2	12	32	36	52	18	8	52	42	28	22	12	22	6	rest	deca	yed	344	86.00
15 Lysimachia vulgaris	..	..	..	...	...	...	..	...	..	..	..	..	...	1	...	...	...	1	0.25
16 Myosurus minimus	..	..	16	160	103	19	2	5	..	1	..	11	11	4	13	...	2	347	86.75
17 Oxalis corniculata	..	..	..	...	...	...	..	3	5	1	..	1	...	...	2	...	...	12	3.00
18 Papaver Argemone	..	..	5	65	116	16	8	34	30	15	4	26	17	...	...	...	...	336	84.00
19 Papaver dubium	..	..	2	216	169	...	..	1	..	..	..	..	...	...	...	...	...	388	97.00
20 Phyteuma spicatum	..	..	..	...	...	...	..	...	..	..	..	..	...	...	...	...	...	0	0.00
21 Plantago major	..	..	..	...	...	...	..	...	..	..	..	..	...	...	...	...	1	1	0.25
22 Plantago media	..	5	8	4	4	2	1	...	..	..	..	1	...	9	1	6	2	43	10.75
23 Polygonum persicaria	..	3	33	15	2	...	..	2	..	..	..	..	...	...	...	...	...	55	13.75
24 Primula elatior	..	..	..	...	...	...	..	...	..	..	..	..	...	...	...	...	...	0	0.00
25 Potentilla argentea	..	..	3	15	57	67	58	74	7	..	..	2	3	3	3	2	7	301	75.25
26 Scrophularia nodosa	..	..	1	5	27	96	42	47	6	..	..	..	...	...	7	...	...	231	57.75
27 Thlaspi alpestre	..	..	..	...	...	...	..	...	..	..	..	..	all	deca	yed	...	...	0	0.00
28 Thlaspi arvense	..	..	..	...	1	...	..	2	..	..	..	2	10	11	11	15	35	87	21.75
29 Verbascum nigrum	..	..	..	...	...	...	..	...	..	..	..	..	...	...	...	...	...	0	0.00
30 Veronica Beccabunga	..	..	..	...	...	...	..	1	1	1	6	25	42	103	...	4	...	183	45.75
31 Veronica officinalis	..	..	..	...	...	8	56	332	..	..	..	..	...	...	rest	deca	yed	396	99.00

cave on the inner. The lower seed, lying nearer the base of the bur, is concave on the outer face and convex on the inner. ARTHUR found that plantings of burs of the cocklebur (mainly *Xanthium canadense*) in a garden bed resulted in the germination of all the lower seeds in the first year after ripening, and of only a very small per cent. of the upper seeds. The second year a great majority of the upper seeds grew. A few of the upper ones, however, grew in the third and fourth years.

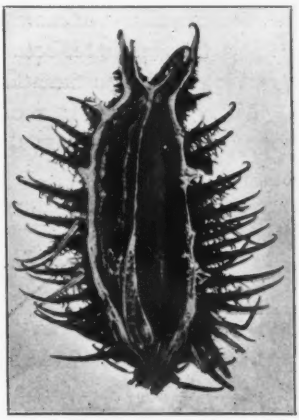


FIG. 1.—Cocklebur cut away to show upper and lower (dimorphic) seeds.

MASTERMAN (**7**) claims that both seeds grow the first year after they are planted. His methods, however, were not at all adapted for detecting whether the two seeds of the bur have markedly different germinative characters. Both ARTHUR'S work and the experimental results of this paper show that such differences in the germinative characters undoubtedly exist.

Delayed germination was found by NOBBE and HÄNLEIN to be due in many cases to the impermeability of the seed coat to water. As to the seeds listed in Table I, however, they say that in every case the seeds absorbed the water readily and yet lay in the germinator in a saturated condition for long periods, either not germinating at all or scattering their germination over a long period. They maintain that in cases where water is admitted the growth after long exposure to germinative conditions must be due to some change going on within the embryo during the period of rest. WINKLER (**15**), WIESNER (**14**), KIENITZ (**4**), and PFEFFER (**9**) expressed similar views.

ARTHUR found that both seeds of the cocklebur take up water readily while in the bur. The bur, therefore, does not account for the delay. He also believes that the extremely delicate seed coats are in no way different in the two seeds and that the structure of the seeds, therefore, offers no explanation of their germinative difference.

He suggests that enzymes are produced readily in the lower seeds and that, therefore, they have foods at hand to begin their growth immediately; whereas the upper seeds are able to develop digestive ferments only after a long period of rest, and hence their germination is delayed one or more years. This theory has its experimental basis in the fact that if both sorts of seeds are exposed to germinative conditions for some time the lower ones show much reducing sugar, while the upper ones have only a trace. My interest in this problem was aroused by ARTHUR'S paper on the cocklebur, and the work was begun for the purpose of testing this enzyme theory and determining definitely the cause of the delayed germination of the upper seed.

II. Materials and methods.

Most of the germinative tests reported in this paper were made between moist filter papers, but in all these cases corresponding tests with very similar results were made in fine quartz sand and in garden soil. For *Avena fatua*, Iris, and cocklebur seeds in the bur, all tests were made in sand and garden soil on account of the great liability of these structures to be attacked by fungi.

Five species of cocklebur were used: *Xanthium canadense* Mill., *X. echinatum* Murr., *X. glabratum* (DC.) Britton, *X. glanduliferum* Greene, and *X. speciosum* Kearney. In each species similar conditions gave similar results whether the seeds were in the bur or removed from it; but for convenience in handling and accuracy of the records the seeds were generally removed from the bur. For testing increased oxygen pressures the soaked seeds were allowed to rest on the walls of flasks containing oxygen or (in the checks) air. Germinative tests at high temperatures were made in ordinary paraffin ovens regulated to the desired temperature. The effect of temperature on the rate of diffusion of oxygen through the seed coats of the cocklebur was determined in a large water bath (such as is used in chemical laboratories for determining solubility, etc.) regulated to 0.01°.

The seeds used were collected when thoroughly ripe from various parts of the United States and Europe,[1] stored in a dry room, and used in experimentation within six months after collected. Since a year of dry storage and the region from which the seeds were

[1] I am indebted to M. P. DIEUDONNÉ for collections from Belgium.

gathered gave no marked germinative variations in the species used, the time of storage and the region in which they were collected need no further consideration. A seed was considered germinated when root hairs appeared, except in Xanthium and Iris, where late development of root hairs made this test worthless and the lengthening of the radicle 5^{mm} was considered the criterion of germination.

III. Experiments.

1. Cocklebur.

Effect of enzymes.—When work was begun on the seeds of the cocklebur, Arthur's enzyme theory was adopted as a working hypothesis, on the supposition that it would involve a study of the difference in the development and action of the enzymes in the two seeds. As Waugh (**13**), Stone (**10**), and Thompson (**11**) were able to increase markedly the germination of old seeds by soaking them in solutions of various enzymes, it was thought that perhaps the upper seeds of the cocklebur could be made to germinate without delay by merely soaking them in solutions of pepsin, or plant trypsin, or in filtered extracts from the germinated lower seeds. Experiments in this line gave only negative results, but led to the discovery that high temperatures cause the immediate germination of some of the upper seeds. The enzyme theory was abandoned after failures to detect any differences in the digestive activities of extracts of the upper and lower seeds. At this point the results showed that the difference in the germinative characters of the two seeds had other causes; hence a new line of experiments was begun.

Effect of high temperatures.—High temperatures bring about the immediate germination of the upper seeds of *X. echinatum.* Table II shows the results of temperature experiments with this species. Results were similar whether soil or filter paper was used for germinators and whether the seeds were in the burs or removed from them.

As Table II shows, the lower seeds of *X. echinatum* germinate readily at 22–24°, but even more readily at 32–34°. The upper seeds do not germinate at all at 22–24°, but respond readily at 32–34°. The lowest temperature at which any considerable per cent. of the upper seeds grow is at 33°, and the similar point for the lower seeds

TABLE II (*X. echinatum*).

Temp. °C.	Seeds	Per cent. germinated after			
		1 day	2 days	5 days	8 days
22–24.....	upper	0	0	0	0
	lower	3	31	87	99
32–34.....	upper	8	55	99	99
	lower	23	100	100	100

is 23°. In *X. canadense*, *X. glabratum*, and *X. speciosum* the upper seeds germinate only sparingly at a constant temperature of 35°, but to a considerably larger per cent. at a temperature fluctuating between 25 and 40°. The lower seeds of *X. canadense* germinate readily at 18–21°, while the lower seeds of *X. glabratum* have a minimum germinative temperature of about 23°. The highest minimum germinative temperature yet reported, 15.6–18.5°, is recorded by Detmer (**2**) for the cucumber and watermelon.

From the above data it may be seen that in the cocklebur there are remarkably high minimum germinative temperatures. *X. echinatum*, the least remarkable of the four species studied in this respect, has this critical temperature 15° higher in the upper seed and 5° higher in the lower one than that of the watermelon and cucumber.

Effect of wounding.—In removing seeds from the burs the knife often clipped off a small portion of the distal ends of the cotyledons. It was observed that the upper seeds so wounded begin a marked growth in the wounded region even at the temperature of 20–22°. The growth gradually moves down the cotyledons until it reaches the radicle. This reverses the normal method of germination. Normally the radicle first pushes out, sets itself in the ground, and lifts the cotyledons above the soil, after which they begin their growth. When the upper seeds are wounded at the radicle end, either by a slight cut or a pin prick, germination takes place in the normal way. This observation suggested complete removal of the seed coat.

Effect of removing the seed coat.—After the seeds have been soaked six hours the extremely delicate seed coats can be removed, without the least injury to the embryo, by merely pinching the seed between thumb and finger. The coat-free upper and lower seeds of any one of the four species studied germinate with almost equal readiness at any

point within their temperature limits, and the two seeds have almost identical temperature limits. Table III shows the relative speed of

TABLE III.

X. canadense; COATS REMOVED; TEMP. 18–22°.

SEEDS	PER CENT. GERMINATED AFTER			
	3 days	4 days	6 days	9 days
Upper.....	47	75	84	100
Lower.....	51	77	89	100

germination of the upper and lower seeds of *X. canadense* at 18–22°. The minimum germinative temperature for the upper and lower seeds of this species with the seed coats removed is about 18°. The same point for other species mentioned is 2–3° higher. Each seed of Xanthium has then two minimum germinative temperatures: one with the seed coat intact, and a lower one with the seed coat removed. In the upper seeds these two temperatures differ by fifteen or more degrees and in the lower ones by two or more degrees. Table IV gives the approximate germinative minimum temperatures for each

TABLE IV.

MINIMUM GERMINATIVE TEMPERATURES.

Species	Seeds	Minimum temp. °C. coats intact	Minimum temp. °C. coats removed
X. canadense.....	upper	fluctuating 25–41	18
	lower	21	18
X. echinatum.....	upper	32–33	19–20
	lower	23	19–20
X. glabratum.....	upper	fluctuating 25–41	20
	lower	23	20
X. speciosum.....	upper	fluctuating 25–41	20
	lower	22	20

of the four species with seed coats intact and seed coats removed.

It is evident from the results so far given that ARTHUR'S statement that the difference between the two seeds does not lie external to the embryo, but in the embryos themselves, is entirely wrong. He overlooked the real point of difference, the seed coats, because of their extreme delicacy and because of the ease with which they admit water.

Effect of increased oxygen pressures.—Since the delay is secured by the seed coat, it must exclude either water or oxygen. As ARTHUR states, both seeds seem to take up water with equal readiness. I found that in eighteen hours of soaking, the upper seeds of *X. canadense* imbibed 51 per cent. of their dry weight, while the lower ones imbibed 62 per cent. In the same time the upper seeds of *X. echinatum* imbibed 48 per cent. and the lower ones 47 per cent. The difference then in water imbibition will not serve to explain the difference in the germinative characters of the two seeds.

It was found best in testing increased oxygen pressures to soak the seeds 12–18 hours and then allow them to stick to the walls of a flask or bottle containing the oxygen or (in the check) air. After being thus treated and kept at 21–23° for six days, the upper seeds of *X. canadense* gave 100 per cent. of germination in pure oxygen and 0 per cent. in air. The growth in the seeds germinated in oxygen at these relatively low temperatures does not take place in the normal way. It begins in the distal region of the cotyledons and works down toward the radicle, as was described for seeds wounded at the distal end of the cotyledons. This peculiarity seems to be related to the structural character of the seed coat.

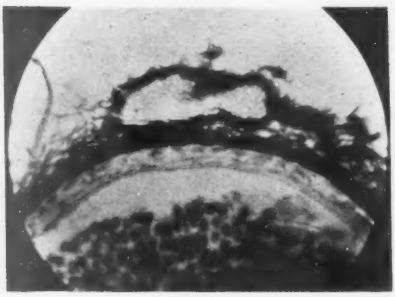


FIG. 2.—Cross section of the seed coat of Xanthium, showing the three layers.

The seed coat consists of three distinct layers (*fig. 2*). The outer layer consists of shell-like cell walls which are more and more collapsed as the inner portion of the layer is approached. This layer is traversed by several groups of tracheae which are parallel with the long axis of the seed. The middle layer is very dense, apparently consisting of collapsed cell walls and staining very deeply with safranin. The inner layer consists of 1–5 layers of cells containing protoplasm and nuclei. Each layer is thickest at the radicle end and gradually becomes thinner toward the distal end of the cotyledons.

Turn to page 288 for the beginning of this discussion

The measurements of the three layers are approximately as follows: the outer layer, at the radicle end 100 μ, at the distal end 20 μ; the middle layer, at the radicle end 15 μ, at the distal end 8 μ; the inner layer, at the radicle end 30 μ, at the distal end 4 μ. The middle layer is somewhat thicker in the upper than in the lower seed. In both seeds it is less dense at the distal end. It seems probable that the denser middle layer is only partly permeable to oxygen, less so in the upper than in the lower seed, and less so at the distal than at the radicle end. This accounts for the increased oxygen pressure initiating the growth at the distal end.

Gaseous exchanges in respiration.—The two sets of experiments on the gaseous exchanges of the seeds in respiration throw some light on the effect of the seed coats. The first set, reported in Table V, shows the ratio of the oxygen taken up with the seed coats removed to that taken up with the seed coats intact. Only the ratios can be compared here, for different weights of seeds were used in the four different determinations. These measurements were made with eudiometers.

TABLE V.

ABSORPTION OF OXYGEN; SEED COATS INTACT AND REMOVED; TEMP. 23°.

SEEDS USED	COATS	CC. OF OXYGEN TAKEN UP AFTER		RATIO OF I TO II	
		6 hrs.	22 hrs.	6 hrs.	22 hrs.
Lower seeds, X. canadense	removed I	2.5	12.6		
	intact II	1.6	6.9	1.6	1.8
Lower seeds, X. echinatum	removed I	12.2			
	intact II	7.1		1.7	
Upper seeds, X. glanduliferum	removed I	4.4	11.0		
	intact II	2.1	3.6	2.1	2.4
Upper seeds, X. echinatum	removed I	9.4	13.4		
	intact II	4.5	6.7	2.1	2.0

As Table V indicates, the lower seeds at 23° take up 1.6 to 1.7 times as much oxygen with the seed coats removed as with the seed coats intact; while the upper seeds take up 2 to 2.4 times as much oxygen with the seed coats removed as with them intact. The ratios are probably much smaller than would obtain in the soil or between wet filter papers, for the coats of the seeds lying on the walls of the eudiometer, though in a theoretically saturated atmosphere,

became relatively dry. As is found by actual measurement, oxygen diffuses through the relatively dry seed coats much more rapidly than through those saturated with water. This is indicated again by the fact that the soaked upper seeds of *X. echinatum* germinate readily at 25–27° when resting on the walls of corked bottles containing air, while in soil or between saturated filter papers a temperature of 33° is necessary for any considerable germination. The upper seeds of every species with seed coats intact can be most easily germinated by allowing the seeds to lie on the walls of corked bottles of such size that a good oxygen supply is given. In this condition the relatively dry coats allow the passage of considerably more oxygen, hence germination comes about at lower temperatures.

The second set of experiments on gaseous exchange is for the purpose of determining why high temperatures bring about the germination of the upper seeds with the seed coats intact. It immediately suggests itself that this may be due to one or both of two things: the amount of oxygen diffusing through the seed coat may rise with the rising temperature, or the amount of carbon dioxide evolved may become greater in proportion to the amount of oxygen consumed as the temperature rises.

It is seen in the 6-hour 50-minute column, as well as in the 12-hour column of Table VI, that the seeds with the coats intact at 19°, whether upper or lower, take up less than half as much oxygen as is taken up by the seeds at 33°. This indicates that the diffusion of oxgyen through the seed coats is much slower at 19° than at 33°. This conclusion, however, needs more direct evidence. Especially is this apparent when it is remembered that 19° is slightly below the minimum germinative temperature, even with the coats removed, and that the amount of oxygen consumed, therefore, may not represent the full amount that can diffuse through the coats at 19°, provided the consumption on the inside is complete.

The apparatus in *fig. 3* was used for determining accurately the effect of high temperatures on the rate of diffusion of oxygen through the seed coats. *S* is a storage bottle for potassium pyrogallate; *f*, a flask from which the oxygen is to be absorbed by potassium pyrogallate; *t*, a seed coat fastened on the end of a glass tube; *w*, a vial of water from which a thread reaches the seed coat to keep it wet; *c*, a capil-

lary tube sealed at the upper end with wax; c', a small graduated tube by which the rate of diffusion of oxygen is to be ascertained; and a, an air chamber. After s is furnished with 300^{cc} potassium pyrogallate the whole apparatus, excepting pinchcock 1 and the portions of tubes c and c', is immersed in a water bath regulated to the desired temperature within .01°. After the apparatus has had sufficient time to attain the temperature of the bath, pinchcocks 1 and 2 are loosened; the plugged end of c filed off; and the pryogallate forced into f. Now c is resealed and the pinchcocks reclamped; a drop of water is allowed to enter the graduated tube c'; and the rate of diffusion of the oxygen is read by the rate of the movement of the drop in tube c'. The same seed coat can be used repeatedly at temperatures to be compared. Numerous measurements made in this way showed the rate of diffusion 1.4 to 1.6 as fast at 33° as at 19°.[2]

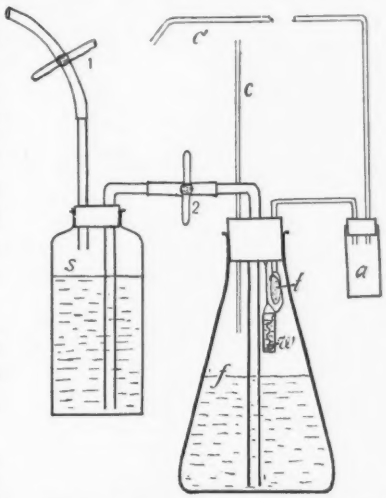


FIG. 3.—Apparatus for testing permeability of seed coats of cocklebur to oxygen. For description see text.

The amount of CO_2 evolved is considered the best measure for the amount of respiration occurring. It is evident from the measurements recorded above, that if the ratio $CO_2:O_2$ remains constant with the rise in the available oxygen due to the rise in temperature, the amount of carbon dioxide evolved will increase from 1 to 1.4–1.6 as the temperature rises from 19° to 33° with the seed coats intact. If it happens, however, that the ratio $CO_2:O_2$ rises in value along with the rise in the rate of the diffusion of oxygen, then the increase in respiration with this rise in temperature will be still more marked.

[2] When the coat was allowed to dry somewhat the rate of diffusion was also greatly increased.

This involves the necessity of making a study of the respiratory ratios at 19° and at 33°, both with the coats intact and removed.

Table VI shows the effect of high temperatures on the respiratory ratios when the seed coats are intact. In this experiment equal weights (2.68gm) of seeds of *X. echinatum* were used in each case. The results are therefore comparable in every way. It is seen that at 33°, up to the 6-hour 50-minute reading the respiratory ratio for both upper and lower seeds is 1. After this reading the ratio falls rapidly, reaching at the 12-hour reading 0.82 in the case of the lower seeds and 0.87 in the case of the upper ones. It was observed that soon after the 6-hour 50-minute reading the radicles began breaking through the seed coats. In a large number of experiments at 33° the respiratory ratio always fell rapidly after the radicles broke the seed coat. At 19°, with the seed coats intact, the respiratory ratio is 0.6 in the lower seeds and 0.64 in the upper ones. Many measurements at 33° with seed coats removed showed a respiratory ratio of 0.7–0.8 in both the upper and lower seeds, while numerous measurements at 19° with seed coats removed always gave a respiratory ratio of about 0.6.

The facts may be summarily stated thus: the respiratory ratio with the seed coats removed is about 0.6 at 19° and 0.7–0.8 at 33°; with the seed coats intact it is 0.6 at 19° and 1.0 at 33°. From these results two conclusions are plain: the respiratory ratio rises considerably (from 0.6 to 0.8) with a rise in temperature from 19° to 33° under the free oxygen supply secured by the removal of the seed coats; but it rises much more (from 0.6 to 1.0) with the same

TABLE VI.

X. echinatum—SEED COATS INTACT; 119 UPPER SEEDS; 88 LOWER; 2.68 GM.

SEEDS	TEMP. °C.	CC. INTERCHANGE OF GASES AFTER											
		4 hrs. 50 min.			6 hrs. 50 min.			10 hrs.			12 hrs.		
		O_2	CO_2	$\frac{CO_2}{O_2}$	O_2	CO_2	$\frac{CO_2}{O_2}$	O_2	CO_2	$\frac{CO_2}{O_2}$	O_2	CO_2	$\frac{CO_2}{O_2}$
Lower	33	3.3	3.3	1.	4.9	4.9	1.	7.9	6.5	.83	9.9	8.3	.82
Upper	33	2.8	2.8	1.	4.1	4.1	1.	6.	5.3	.88	7.1	5.9	8.7
Lower	19				1.8	1.1	.61				4.0	2.4	.60
Upper	19				1.5	.94	.63				3.3	2.1	.64

rise in temperature when the oxygen supply is diminished by the presence of the seed coats.

This rise in the respiratory ratios with the rise in temperature is measured for the entire seed (more than 95 per cent. of which is storage material) and not for the actively growing radicle. It is probable that for the radicle the respiratory ratio rises far above 1. This becomes more evident when it is remembered that at 33° the radicle grows first, as is normal; but that at 22°, in increased oxygen pressures, though it be five atmospheres, the growth of the cotyledons first takes place. Here, too, it should be recalled that the seed coat is much thicker and more dense over the radicle than over the cotyledons.

From the data of this section two effects of a rise in temperature are evident. 'It increases the rate of diffusion of oxygen through the seed coats; and it increases the respiratory ratio somewhat with the seed coats removed and markedly with the seed coats intact. If CO_2 be taken as the criterion, it is possible from the data above to calculate quantitatively the increase in respiration with a rise in temperature from 19° to 33°, when the seed coats are intact. The increase in the rate of diffusion of oxygen (if there were no increase in the ratio $CO_2:O_2$) equals an increase in respiration from 1 to 1.5 (the average of 1.4–1.6). But the increase in the respiratory ratio is from 0.6 at 19° to 1.0 at 33°. This then increases the respiration from 6 units to 10 units. When the two facts that indicate an increase in respiration are considered together, it is evident that a rise in temperature from 19° to 33° with the coats intact causes a rise in the amount of respiration from 0.6 to 1.5, or from 1 unit to 2.5 units.

It is evident from Table V that the seed coats of the lower seeds, as well as those of the upper, greatly restrict the amount of oxgyen used by the seeds, and that this restriction, though considerable, is not markedly greater in the upper seeds than in the lower ones. In *X. echinatum* this rather slight difference in the rate of diffusion of oxygen through the seed coats of the upper and lower seeds is yet sufficient to give the upper seeds a minimum germinative temperature of 32° with seed coats intact, and the lowers 22°; while both seeds with the seed coats removed have a minimum germinative

temperature of 19°. Since the difference of the two seed coats in the matter of oxygen diffusion is rather slight, it is not remarkable that the structural difference is not radical. This slight difference, however, is sufficient to raise the minimum germinative temperature and secure the delay of the upper seed.

Growth of upper seeds.—It is now obvious how the *delay* of the upper seeds is secured. But why do they grow at all in nature? This comes about by a partial disintegration of the seed coats, which is clearly shown by a change in appearance from shiny brown to a dull black or in some cases to colorless, and results in the admission of more oxygen. The length of the delay depends upon the ability of the seed coat protected by the surrounding bur to resist the factors of disintegration in the soil. The portion of the bur covering the lower seed decays within a few months after burial, while the portion covering the upper seed is always far more persistent. A variation in the persistence of the portion of the bur covering the upper seed, as well as the variation in the ability of the seed coat to resist the factors of disintegration independent of the bur, gives considerable variation in the length of delay of the upper seed. These facts show why only a few of the upper seeds grow the first year after ripening, the vast majority the second year, and a few not until the third and fourth year.

Table VII shows the effect of a period in the ground upon the seed coats and upon the vitality of the embryos of the upper seeds of *X. canadense*. Burs produced in 1904 were gathered in November of that year and stored in the laboratory until March 1905. At this time half these burs were buried and the other half kept in the laboratory. In November 1905 the upper seeds of 1904 burs stored in the laboratory, of 1904 burs buried eight months, and of 1905 burs gathered from the same patch, were removed and put to germinate. At 28–33°, with coats intact as Table VII shows, upper seeds of 1904 buried eight months gave 96 per cent. germination, upper seeds of 1904 stored in the laboratory gave 0 per cent., and upper seeds of 1905 gave 3 per cent. As shown in the same table, similar seeds with the coats removed in a germinator at 18–22° (near the minimum germinative temperature with the seed coats broken) gave in upper seeds of 1905 prompt germination, in upper seeds of 1904 stored

TABLE VII.

X. canadense; UPPER SEEDS.

SEEDS	COATS	TEMP.	PER CENT. GERMINATED AFTER		
			3 days	6 days	33 days
1905 just gathered	on	28–33	0	0	3
1904 stored in laboratory 1 yr.	on	28–33	0	0	0
1904 in lab. 4 mos., buried 8 mos.	on	28–33	44	88	96
1905 just gathered	off	18–22	48	100	100
1904 stored in lab. 1 yr.	off	18–22	20	42	100
1904 in lab. 4 mos., buried 8 mos.	off	18–22	0	4	94

in the laboratory less prompt germination, in upper seeds of 1904 buried eight months much less prompt germination. From this table two things are evident. A period in the ground causes a partial disintegration of the seed coats which lowers the germinative minimum temperature with the seed coats intact. This accounts for the results in Table VII with the temperature at 28–33° and the coats intact. The vitality (if we mean by vitality the readiness with which seeds will germinate at a given temperature) of the embryos falls somewhat with a year of dry storage and markedly with eight months in the ground.

2. AXYRIS AMARANTHOIDES.

L. R. WALDRON of the North Dakota Agricultural College informed me that *Axyris amaranthoides* bears two kinds of seeds. One grows soon after being subjected to germinative conditions and the other fails to grow under similar conditions. The former, which is flattened and winged (*fig. 4, a*) he designated as *a*; the latter, which is almost spherical (*fig. 4, b*), as *b*. From material kindly furnished by him I have found that the distal portions of the branches bear entirely form *a*, and the proximal portions entirely form *b;* while the intermediate zone bears both forms even within the same seed cluster. Every seed is either one form or the other, there being no intergrading. Seeds of form *a* and *b* are about equal in number. It is found that form *b* fails to grow because the seed coat is only very slowly permeable to water. Form *a* soaked in water at 23°

FIG. 4.—Dimorphic seeds of *Axyris amaranthoides.*

absorbed after 24 hours 39 per cent. of dry weight, after 48 hours 70 per cent. Form *b* absorbed after 24 hours 4 per cent., after 48 hours 5 per cent., after 120 hours 6.5 per cent., and after 174 hours 9.5 per cent. After 174 hours soaking, 2 per cent. of form *b* had swollen up and germinated. At 23° with seed coats broken, as shown in Table IX, form *b* germinates much more readily than form *a* with seed coats intact, and somewhat more readily than form *a* with seed coats broken. It is evident from these tests that the embryo of form *b* is more vigorous than the embryos of form *a*.

It should be pointed out that the embryo of form *b* has an ideal storage condition. The water-excluding seed coat keeps it dry as it lies buried in the ground with the temperature relatively low. With the partial disintegration of the coat comes the admission of water and the growth of the embryo. The length of the delay in germination thus secured will vary greatly with water and temperature conditions, and with the different individuals of form *b*. It is probable that it will amount to many years in some cases.

In Axyris, as in the cocklebur, the same plant bears two sorts of seeds. One sort grows rapidly in nature and the other only after a considerable delay. Unlike the cocklebur, the seeds are not paired, and the delay is secured by the seed coat shutting out water rather than oxygen.

3. Abutilon Avicennae and Chenopodium album.

Agriculturists claim that the seeds of *Abutilon Avicennae* lie in meadows and pastures for twenty years without growing, but upon breaking the soil grow in great abundance. When these seeds are soaked in water for forty-eight hours about 13 per cent. swell up, while the embryos of 87 per cent. remain extremely dry and can be pulverized. After weeks of soaking only a small per cent. additional will swell, a few at a time. For the relative per cent. of germination of these seeds with seed coats intact and seed coats broken see Table IX.

In *Chenopodium album*, mentioned in the Nobbe-Hänlein table, about 16 per cent. of a crop of seeds swell up after twenty-four hours soaking. By continual soaking the remaining seeds gradually swell a few at a time, but much more readily than is the case with Abutilon.

Contrary to NOBBE AND HÄNLEIN's conclusion, the distributed germination shown by this species is secured by the slowness with which water penetrates the seed coats.

While in Axyris and Xanthium delayed or distributed germination is secured by peculiar seed coat characters of one form of the dimorphic seeds, in *Abutilon Avicennae* and *Chenopodium album* the distributed germination is secured by a variation in the seed coat characters of similar seeds.

4. IRIS.

Dr. C. J. CHAMBERLAIN informed me that he had never succeeded in germinating seeds of various species of Iris, although he had often attempted it in order to have root tips for cytological purposes. The bulk of the seed consists of the horny endosperm with food stored on the walls as hemicellulose. On one side of the endosperm is a cylindrical cavity in which the embryo is borne. The cavity is covered by a cap, thus entirely closing in the embryo. When the seed is dry, the embryo only partially fills the cavity, but after twenty-four hours soaking it completely fills it. In this condition, however, the seeds will lie for weeks without germinating. If now the caps are removed and the seeds still kept in the water, the embryo protrudes 3–7^{mm} within an hour. Seeds with caps removed germinate very readily, while those with caps intact do not germinate at all. For the effect of removing the cap in *Iris sibirica* and *I. Pseudacorus*, see Table IX. Increased oxygen pressure and high temperatures with the caps intact did not cause germination. With the caps removed, the most successful germination was secured by using sterilized sand as a germinator at 28–33°.

The amount of moisture absorbed by the embryo within the limiting wall of the endosperm is not sufficient to permit growth to begin. By taking away this limit to water absorption by removal of the cap or a portion of the endosperm in the region of the embryo, absorption is resumed and growth soon begins. Judging from a number of observations, it appears that in nature long soaking and accompanying disintegration lead to the loosening of the cap, or more frequently to the decay of the endosperm at one side of the embryo.

TABLE VIII.

Species	Temp. °C.	Coats	Per cent. germinated after				
			2 days	3 days	5 days	9 days	38 days
Plantago major	28–33	entire	20	28	44	54	60
		broken	50	81	92	96	96
	18–22	entire	0	0	0	0	0
		broken	0	0	0	0	0
Plantago Rugelli	28–33	entire	2	2	9	15	38
		broken	91	98	98	98	98
	18–22	entire	0	0	0	0	0
		broken	0	0	0	12	12
Thlaspi arvense	28–33	entire	32	38	41	41	41
		broken	94	100	100	100	100
	18–22	entire	0	0	0	0	0
		broken	48	75	86	93	96

I was obliged to abandon work on these seeds, for on account of handling them I had repeated and severe attacks of dermatitis from contact with the syrupy covering of the endosperm. The symptoms were identical with those of Rhus poisoning.

5. The Nobbe-Hänlein Table.

Beside *Chenopodium album*, I have studied the following seeds mentioned in the Nobbe-Hänlein table: *Aquilegia vulgaris*, *Capsella Bursa-pastoris*, *Lysimachia vulgaris*, *Plantago major*, *P. Rugelii*, and *Thlaspi arvense*. It was found, in agreement with Nobbe and Hänlein, that all these seeds absorb water readily.

In *Aquilegia vulgaris* Nobbe and Hänlein obtained a germination of only 0.75 per cent. after sixteen days, and no more during the remaining three years. In all tests at 23° I found over 50 per cent. germinating within thirty days. None generally germinated short of sixteen days because of the rudimentary state of the embryo. Breaking the seed coats cut down the percentage of germination by allowing infection by fungi which the slow growing embryos were unable to resist.

Table VIII shows the germination of seeds of *Plantago major*, *P. Rugelii*, and *Thlaspi arvense* at 18–22° and 28–33° with seed intact and seed coats broken. It is seen that with seed coats broken and with favorable temperatures over 95 per cent. germinate in every case. These results should be compared with Table I, in which

NOBBE and HÄNLEIN show the germination of only 0.25 per cent. of *P. major*, 10.75 per cent. of *P. media*, and 21.75 per cent. of *Thlaspi arvense* after 1,173 days. In the last species the germination is late in that period.

From Table VIII several important facts are evident. The temperature of 18–22° is below the minimum germinative temperature of seeds of *P. major* with seed coats intact or broken, and below that of *P. Rugelii* and *Thlaspi arvense* with seed coats intact. It is very near the germination minimum of *P. Rugelii* with seed coats broken and well above that of *Thlaspi arvense* in similar condition. A very marked increase in germination is secured by rupturing the coats even when the most favorable temperatures are used. High temperatures, then, will overcome only in part the seed coat effects. High temperatures are more efficient in overcoming the seed coat effects in *P. major* than in *P. Rugelii*.

In Table IX a similar effect of rupturing the seed coat is shown for *Capsella Bursa-pastoris*, *Lysimachia vulgaris*, and *Euphorbia Cyparissias*. It should be mentioned that the region and extent

TABLE IX.

TEMP. 18–22° EXCEPT FOR IRIS, WHICH WAS 28–33°.

SPECIES	COATS	PER CENT. GERMINATED AFTER					
		1 day	3 days	6 days	14 days	20 days	30 days
Abutilon Avicennae	entire	0	13	13	13	13	13
	broken	48	98	98	98	98	98
Avena fatua	entire	0	0	8	8	8	8
	broken	0	4	92	96	96	96
Capsella Bursa-pastoris	entire	0	8	14	15	15	15
	broken	0	80	100	100	100	100
Chenopodium album	entire	0	10	16	16	16	16
	broken	0	80	100	100	100	100
Euphorbia Cyparissias	entire	0	0	2	20	20	20
	broken	0	20	84	84	84	84
Lysimachia vulgaris	entire	0	0	0	0	0	0
	broken	0	44	60	64	64	64
Axyris amaranthoides A	entire	0	76	92	94	96	96
	broken	14	100	100	100	100	100
A. amaranthoides B	entire	0	0	0	0	0	0
	broken	55	100	100	100	100	100
Iris sibirica	cap on	0	0	0	0	0	0
	cap off	0	6	18	75	98	98
Iris Pseudacorus	cap on	0	0	0	0	0	0
	cap off	0	14	32	87	97	97

of the rupture makes no difference so long as the embryo is not injured.

NOBBE and HÄNLEIN make no mention of the temperature maintained in the course of their experiments and seem unconscious of the fact that the temperature plays an important part. Judging from the results in Table VIII as compared with their results in Table I, they must have run their germinators at relatively low temperatures. These investigators, as well as WINKLER, were likewise entirely unaware of the effect of the seed coats upon germination.

6. AVENA FATUA.

Avena fatua has some germinative characters which are interesting and which show that the seed coat characters just described for other seeds appear in the grasses. Table IX shows that at 18–22°, 8 per cent. grow with seed coats entire and 96 per cent. with coats broken. At 33°, 50–60 per cent. grow with coats entire and 97 per cent. with the coats broken. This seed coat character probably distributes the germination of a given crop over a period of years. It probably accounts for the claim of farmers that these grains will lie in pastures and meadows for twelve to fifteen years and then grow abundantly when land is plowed. WALDRON (**12**) believes this idea is wrong, but it is easy to see how his vitality tests might be entirely misleading, for the seed coat character just described was not taken into account.

7. HAWTHORNS.

I found, as is popularly believed, that no hawthorn seeds will grow immediately after ripening. The seeds of various species were tested by removal of seed coats and subjection to high temperatures and high oxygen pressures; but none of these conditions sufficed to cause germination. Seeds that lay in the soil for a year or more germinated to some extent; while seeds stored in the dry for a similar period did not germinate at all, although the tests were made with naked embryos as well as with seeds bearing the coats. It is evident, therefore, that the change that must precede germination is in the embryo itself rather than in the seed coat; but it is also more or less a matter of disintegration, as is true in seeds whose germination is delayed by seed coats.

IV. General considerations.

Two statements of ARTHUR concerning the cocklebur need special consideration. He says: "Seeds in the bur retain their germinative power, when kept in a dry room, for two years or more; but seeds removed from the bur dry out within a few days and will no longer grow. Seeds removed from the bur and placed in a germinator retain their bright polished appearance as long as they are alive; when dead they turn dull and lusterless." I find that the seeds retain their vitality fully as well when removed from the bur and allowed to dry as when in the bur. In fact, a dry cool place is the best for storage of these seeds whether in or out of the bur, as is true for most seeds. The seeds removed from the bur and kept in a dry place retain their vitality much more than five years. I found the condition of the seed coat no indicator of the vitality of the embryo. The coat through disintegration loses its luster and turns black or sometimes colorless, which means that more oxygen is admitted and that the minimum germinative temperature of the seed has fallen. It is not surprising that ARTHUR drew these conclusions, for his work gave him no idea of the germinative conditions of the upper seeds or of the significance of the seed coat.

As DUVEL (**3***a*) states, seeds retain their vitality longest in conditions that permit of least respiration. KOLKWITZ (**5**) has shown that respiration is extremely slight in dry seeds at low temperatures. The embryos of seeds whose germination is delayed by coats that exclude oxygen, such as Abutilon, Axyris, and Chenopodium, are kept very dry by the coats. As they lie in the ground they are likewise relatively cool. In nature, in short, they have the most favorable storage conditions up to the time when the coats, through partial decay or long exposure to water, admit moisture and germination begins. It is not wonderful that such seeds lie in the ground twenty to twenty-five years and yet retain their vitality. While the reduction in the oxygen admitted to the upper seed of the cocklebur cuts down the respiration considerably, it does it to no such extent as does the exclusion of water. The coats that exclude water are undoubtedly much better adapted to securing a long delay than are the coats that merely exclude oxygen. In nature the longer delays are certainly secured by the former method.

PFEFFER (9) says: "The conditions which lead to certain seeds resting under the soil for as long as fifty years and germinating when dug up have not as yet been determined." This, as well as the sudden appearance of weeds in forests after fires and in meadows of many years standing immediately upon plowing, is probably explained by a few simple facts. Weed seeds are produced in great abundance. Because of variation in seed coat characters or in some cases of embryo characters, a given crop distributes its germination over a period of years. Seeds deep in the soil germinate less readily because of lack of oxygen or water, and those that do grow exhaust the stored food before reaching the surface. Bringing such seeds to the surface greatly increases their germination and removes the danger of exhaustion of the stored food. The plants of meadows and forests keep the water supply reduced and thereby cut down the chances for the germination and later growth of the weed seeds present. With the destruction of the plants of the forest or meadow comes a great increase in the germination of the weed seeds and a removal of the opposition to their future growth. These phenomena, then, will probably all be explained by a study of the germinative characters of the seeds such as is described in the experimental portion of this paper, along with certain other well established facts on germination.

It is undoubtedly true that many of the tests that have been made for the vitality of weed seeds are untrustworthy, because the significance of the seed coats has been overlooked. This is clearly shown by the results of DUVEL (3*b*) and WALDRON (12), who have carried on extensive experiments to determine the length of time weed seeds must be buried in order to lose their vitality. In column I of Table X is shown the percentage germination determined by me with seed coats broken and with favorable temperatures. In column II appear DUVEL'S results, in which he uses what he terms the "most favorable temperatures," but overlooks of course the seed coat effects. The figures quoted from DUVEL are from the column "original samples," which means fresh seeds, as were the seeds for determining the percentages of column I.

The effect of rupturing the seed coats, as is shown in this table, is very evident, although DUVEL has in part overcome the seed coat effects by high temperatures. The average percentage of germina-

TABLE X.

	I	II
1. Axyris amaranthoides	100	0
2. Xanthium pennsylvanicum	98	50
3. Thalaspi arvense	100	57
4. Plantago Rugelii	96	4
5. Avena fatua	96	75
6. Plantago major	96	24
7. Chenopodium album	100	67

tion for the seven species tested is 98 per cent. with seed coats ruptured, 40 per cent. with seed coats intact. It may be urged that the seeds used by DUVEL are of low vitality. This, however, does not seem at all probable, for I obtained only slightly higher percentages, as shown in Tables VIII and IX, with the coats intact and with favorable temperatures, than those reported by DUVEL. These slightly higher percentages can be accounted for by the fact that the temperature used by me was 28–33°, while the temperature used by DUVEL was 20–30°. Two species of seeds mentioned in this table need special consideration. In *Axyris amaranthoides*, DUVEL determined the vitality as 0 per cent. This is exactly what would be expected if form *b* alone (as shown in Table IX) were used, and if the effect of the seed coat were overlooked. In *Xanthium pennsylvanicum* he finds 50 per cent. germinating. This, too, is what would be expected if the upper and lower seeds of the cocklebur were put in a germinator at 20–30°. The lower seeds would germinate in this condition and the others fail to do so. It seems probable, then, that in Xanthium and Axyris DUVEL overlooked the dimorphic character of the seeds, as well as the effect of the coats on germination. Vitality tests of this kind, that neglect the effect of the seed coats, are tests of the condition of the seed coats rather than tests of the real vitality of the embryos themselves. It is evident that if these errors appear in the original tests for vitality they will likewise appear in the tests made after different periods of burial. If vitality tests, looking to the extermination of weeds, are to be of real value, the exact germinative character of each species must first be determined, and all vitality tests must then be made on the basis of these germinative characters.

It is obvious that the seeds which fail to grow in the ordinary grain tests often do so because of seed coat characters rather than because of lack of vitality of the embryos. This, however, does not in any wise invalidate the ordinary methods of testing grains to be used for seeding, since seeds that are delayed a month or more in germination are of no value in producing the crop. On the other hand, when it comes to testing weed seeds, looking toward extermination, it is highly important that these seed coat characters be taken into consideration.

I am impressed by the high vitality of weed seeds. This is especially true of the more noxious weeds and those in which the seed coat secures a long delay. The high vitality is not shown alone by a quick response to germinative conditions. The percentage of germination in noxious weeds, provided real germinative conditions are given (the seed coat hindrance removed), is very close to 100; and a marked growth of the embryo generally takes place within two days after being subjected to germinative conditions. After recognizing this fact, one is led to suspect that many other cases of low vitality in weed seeds mentioned by DUVEL and others (not examined in this paper) must be due to seed coat characters rather than to lack of vitality in the embryos.

While this paper indicates, exactly contrary to the conclusions commonly held, that delayed germination is in most cases secured by seed coat characters, it yet recognizes that in the hawthorns delay is secured by embryo characters. It is probable that a number of other seeds will be found to belong to the same category as the hawthorn. It is of great interest to know just the changes which take place in the seeds of the hawthorns and finally lead to germination through long exposure to germinative conditions. This subject is now under investigation. It must be urged that, until these changes are understood, any attempt to determine the vitality of such seeds is futile.

The methods by which seed characters that secure delayed germination have come about naturally deserves consideration. It may be adaptation coming through natural selection, but an attempt to prove this would end in failure. This delay in many cases, however, is of undoubted advantage to the species. ARTHUR urges that in the cocklebur the two seeds are borne in an indehiscent structure,

the bur, and that it is impossible to have the two seeds distributed in space, so a distribution in time is substituted. Why such an indehiscent involucral structure should be developed instead of such a bur as appears in the burdock needs answer. With the indehiscent bur already in existence the advantage is plain. It is clear that such germinative characters as appear in the seeds of *Axyris amaranthoides*, *Abutilon Avicennae*, etc., insure that the soil will always be supplied with these seeds in process of germination. The destruction of existing vegetation, by fire or otherwise, is followed by a quick appearance of these weeds. In species where none of a given crop of seeds grow until a year or more after falling, it would seem that the adaptive characters, if they be such at all, had overstepped the line of greatest advantage.

V. Summary.

1. Delayed germination is reported in the seeds of many plants and, exactly opposite to the common view, its cause generally lies in the seed coats rather than in the embryos; but in the hawthorns, as perhaps in some other seeds, it is due to embryo characters.

2. In the upper seed of the cocklebur the delay is secured by the seed coat excluding oxygen, while in *Axyris amaranthoides*, *Abutilon Avicennae*, and many other seeds, it is secured by the coats excluding water.

3. In Iris seeds the failure to germinate is due to the endosperm and cap stopping water absorption before the quantity necessary for germination is obtained by the embryo.

4. In *Plantago major*, *P. Rugelii*, *Thlaspi arvense*, *Avena fatua*, and others, the real method by which the coats secure the delay is not yet determined, but there is no doubt that the delay is due to the coats.

5. Seed coats which exclude water are much better adapted to securing delays than are seed coats which exclude oxygen, because of the much greater reduction of respiration in the first case.

6. In nature growth of the delayed seeds comes about through the disintegration of the seed coat structures by a longer or shorter exposure to germinative conditions, and the length of the delay depends upon the persistence of the structure securing it.

7. In the cocklebur the bur aids in preserving the seed coat of the upper seed by being most persistent over it.

8. Even in the hawthorns, where the delay is secured by embryo characters, the germination finally comes about in the course of long exposure to germinative conditions and not in dry storage.

9. In the cocklebur the seed coats of both the upper and lower seeds cut down the oxygen supply, but the first the more markedly. This gives the upper seed a much higher minimum germinative temperature and the lower seed a somewhat higher one. Hence we have in the cocklebur seeds two minimum germinative temperatures; one with the seed coats intact and a much lower one with the coats removed. In the upper seeds these differ by fifteen or more degrees; in the lower seeds by three to five degrees.

10. High temperatures bring about the germination of the upper seeds of the cocklebur with coats intact by increasing the rate of diffusion of oxygen through the seed coat and by raising the respiratory ratio.

11. The minimum germinative temperatures of the seeds of the cocklebur, *Plantago major*, *P. Rugelii*, *Thlaspi arvense*, and various other seeds, with the seed coats intact, is far above the highest minimum germinative temperature reported; while in the cocklebur and *Plantago major* with coats removed this critical temperature is considerably above the highest reported.

I am indebted to Professor CHAS. F. HOTTES, of the University of Illinois, for suggesting the problem in reference to the cocklebur, and to Professors JOHN M. COULTER and C. R. BARNES for kind suggestions and assistance during the progress of the work.

LITERATURE CITED.

1. ARTHUR, J. C., Delayed germination in the cocklebur and other paired seeds. Proc. Soc. Prom. Agric. Science **16**: 70–79. 1895.

2. DETMER, W., Keimungsprocess der Samen 427. 1880.

3*a*. DUVEL, J. W. T., Vitality and germination of seeds. U. S. Dept. Agric., Bur. Plant Ind., Bull. 58. 1904.

3*b*. ———, Vitality of buried seeds. Idem, Bull. 83. 1905.

4. KIENITZ, M., Ueber Ausführung von Keimproben. Bot. Centralbl. **1**: 52–53. 1880.

5. KOLKWITZ, R., Ueber die Athmung ruhenden Samen. Ber. Deutsch. Bot. Gesells. **19**:285–287. 1901.
6. KUNTZE, RICHARD E., Germination and vitality of seeds. Published by the Torrey Bot. Club. 1901.
7. MASTERMAN, E. E., Sprouting of cocklebur seeds. Ohio Nat. **1**:69–70. 1901.
8*a*. NOBBE, F., and HÄNLEIN, H., Ueber dei Resistenz von Samen gegen die aüsseren Factoren der Keimong. Landw. Versuchs-Stat. **20**:63–96. 1877.
8*b*. ———, Ueber die Keimkraft von Unkrautsamen. Landw. Versuchs-Stat. **25**:465–470. 1880.
9. PFEFFER, W., Physiology of plants (Eng. ed.) **2**:208. 1900.
10. STONE, G. E., Influence of chemical solutions upon the germination of seeds. Rept. Mass. Agr. Exp. Sta. **13**:74–83. 1901.
11. THOMPSON, A., Zum Verhalten alter Samen gegen Fermentlösung. Gartenflora **45**:344. 1896.
12. WALDRON, L. R., Buried weed seeds. N. Dak. Agr. College Bull. 62. 1904.
13. WAUGH, FRANK A., The enzymic ferments in plant physiology. Science N. S. **6**:950–952. 1897.
14. WIESNER, J., Ueber die Ruheperiode und von einige Keimungsbedingungen der Samen von *Viscum album*. Ber. Deutsch Bot. Gesells. **15**:503–515. 1897.
15. WINKLER, A., Bermerkungen über die Keimpflanzen und die Keimfähigkeit des Samen von *Tithymalus Cyparissias*. Ber. Deutsch. Bot. Gesells. **1**:452–455. 1883.

UNDESCRIBED PLANTS FROM GUATEMALA AND OTHER CENTRAL AMERICAN REPUBLICS. XXVIII.[1]

JOHN DONNELL SMITH.

Saurauja Maxoni Donn. Sm.—Glaberrima. Folia nitida obovato-oblonga in apicem angulo obtuso in basin angulo acuto desinentia calloso-denticulata, nervis lateralibus remotissimis arcuatis. Paniculae subterminales longe pedunculatae folia subaequantes. Sepala paene glabra.

Frutex 3–4-metralis, ramulis laevibus. Folia crassa 14–20^{cm} longa 6–8^{cm} lata, margine vix ac ne vix undulato, nervis lateralibus haud parallelis 1.5–2^{cm} inter se distantibus, petiolis validis 2–3^{cm} longis. Pedunculi 10–17^{cm} longi et panicularum axes complanati striati nitidi, axibus primariis alternis 1.5–2.5^{cm} longis, pedicellis 4–5^{mm} longis, bracteis lincaribus 4^{mm} longis et bracteolis ciliolatis, floribus 2.5^{cm}-diametralibus. Sepala ovalia 7–8^{mm} longa juxta marginem basinque intus pubescentia. Petala discreta oblongo-ovata bilobulata. Filamenta basi cinereo-barbata antheris aequilonga, loculis dimidio discretis. Ovarium glabrum 5- vel rarius 3-loculare, stylis discretis 4.5^{mm} longis.

Secanquiḿ, Depart. Alta Verapaz, Guatemala, alt. 550^{m}, Jan. 1905, *Maxon et Hay* (n. 3221).

Saurauja ovalifolia Donn. Sm.—Strigillosa. Folia ovalia bis longiora quam latiora apice obtusa basi rotundata supra scabriuscula subtus nervatione pubescentia mucronulis denticulata. Paniculae pauciflorae. Sepala strigillosa et pubescentia. Bacca magna villosa.

Frutex orgyalis. Ramuli petioli pedunculi panicularum axes strigillis ferrugineis pilo albo apiculatis operti. Folia 20–23^{cm} longa, nervis lateralibus utrinsecus 20–22, venis utrinque manifestis, petiolis circiter 2.5^{cm} longis. Pedunculi ex quaque axilla nascentes 7–10^{cm} longi, paniculis 4–6^{cm} longis, axibus plerumque geminatis 1–2^{cm} longis, pedicellis 5–10^{mm} longis, bracteolis linearibus 7–10^{mm} longis. Sepala acuminato-ovata 7^{mm} longa. Bacca sordide villosa 9^{mm}-diametralis 5-locularis polysperma, stylis discretis 5^{mm} longis, seminibus obovatis scrobiculatis.—Petala et stamina in exemplis suppetentibus deficiunt. Río Navarro, Prov. Cartago, Costa Rica, alt. 1100^{m}, Mart. 1894, *J. Donnell Smith*, n. 4746 ex Pl. Guat. etc. quas ed. Donn. Sm.

Saurauja subalpina Donn. Sm.—Furfuraceo-strigillosa. Folia longe petiolata oblanceolata utrinque acuta mucronulis incurvis

[1] Continued from BOT. GAZETTE 40:11. 1905.

denticulata supra tuberculis rubiginosis setuligeris scabra subtus nervis approximatis et venis rufo-strigillosa et parce pubescentia. Pedunculi petiolis parum longiores, floribus inter majores. Sepala furfuracea tuberculato-setulosa. Ovarium glabrum.

Ramuli petioli pedunculi panicularum axes simul cano-furfuracei et longe rufo-strigillosi. Folia 20–22^{cm} longa 6–7^{cm} lata, nervis 5–8^{mm} inter se distantibus, venis minutissime reticulatis subtus tantum manifestis, petiolis 3–4^{cm} longis. Pedunculi axillares 4–5^{cm} longi, paniculis circiter 6^{cm} longis floribundis, axibus alternis, pedicellis 4–8^{mm} longis, bracteolis linearibus 3–5^{mm} longis, floribus 2.5^{cm}-diametralibus. Sepala ovata 6–8^{mm} longa, setulis validis brevibus puberulis. Petala discreta obovato-oblonga. Filamenta pilis articulatis barbata antheris infra medium affixis paulo longiora, loculis triente discretis. Ovarium 4–5-loculare stylis discretis 6^{mm} longis bis superatum.

Volcán de Agua, Depart. Zacatepéquez, Guatemala, alt. 3300^{m}, Apr. 1890, *J. Donnell Smith*, n. 2171 ex Pl. Guat. &c. quas ed. Donn. Sm.

PRUNUS CAPOLLIN Zucc., var. **prophyllosa** Donn. Sm.—Ramuli racemiferi peruliferi subaphylli pedunculum lateralem mentientes, perulis ovalibus 12–18^{mm} longis apice retusis sicut stipulae linearilanceolatae glandulo-denticulatae 16–19^{mm} longae atque bracteolae oblongae 7–10^{mm} longae purpurascentibus pubescentibus. Flores nondum satis evoluti solum cogniti.

San Rafael, Depart. Zacatepéquez, Guatemala, Mart. 1905, *Maxon et Hay* (n. 3666).

Tibouchina paludicola Donn. Sm. (§ DIOTANTHERA Triana.)—Patentim glandulari-setulosa. Folia ovata vel lanceolato-ovata acuminata basi rotunda vel leviter cordata supra sparse, subtus nervis venisque, setulifera denticulata 7–9-nervia. Flores axillares singuli vel cymulis trifloris terminales. Stamina glaberrima cum staminodiis alternantia, connectivo longe producto. Ovarium nudum.

Rami subteretes fistulosi et petioli pedicelli calyces setulis patulis glanduliferis conspersi. Folia membranacea 4–8^{cm} longa 2–4^{cm} lata subtus minutius setulifera, petiolis 1–1.5^{cm} longis. Cymulae semel bisve trichotomae foliaceo-bracteolosae, pedicellis lateralibus 5^{mm} longis, dichotomali brevissimo. Calycis setulae purpureae, tubus oblongo-campanulatus 5–6^{mm} longus segmenta attenuato-linearia subaequans. Stamina 5 subaequalia 16–17^{mm} longa, antherarum tubo apicali 2^{mm} longo, connectivo usque ad 3.5^{mm} producto. Staminodia filiformia 4^{mm} longa ananthera. Ovarium ovoideum 3^{mm} altum vertice nec hispidum nec setosum, stylo 2^{cm} longo. Fructus globoso-ovalis 8^{mm} longus.—Species staminodiis et ovario anomala.

In paludibus secus Río Cañas Gordas, Comarca de Puntarenas, Costa Rica, alt. 1100^{m}, Mart. 1897, *Pittier* (nn. 11055, 11056).—In filictis paludosis secus Río El General, Diquis, Comarca de Puntarenas, C. R., alt. 700^{m}, Febr. 1898, *Pittier* (n. 12150).

Monolena Guatemalensis Donn. Sm.—Rhizoma glabrum elongatum. Folia longe petiolata cordato-ovata acuminata integra 7–9-plinervia, lobis basalibus imbricatis. Scapus basi squamis fultus ceterum nudus petiolo longior pluriflorus. Styli basis ovoidea quam capsula major.

Rhizoma 8–17^{mm} crassum 7^{cm} et ultra longum fibrillosum squamis triangularibus 1^{mm} longis ferrugineis conspersum, squamis scapum fulcientibus imbricatis triangularibus vel lanceolatis 3–7^{mm} longis. Folia ob folium alterum caducum quasi alterna 10–16^{cm} longa 6–10^{cm} lata utrinque punctulata et lineolata mucrunculis hinc inde ciliata, petiolis 6–19^{cm} longis. Scapus 14–20^{cm} longus sicut petiolus in sicco rubescens, cyma 4–8-flora, pedicellis 2–3^{mm} longis, bracteis caducis. Calycis florentis tubus carnosulus 3^{mm} altus, lobi membranacei ovati 1.5^{mm} longi. Calycis fructiferi aucti 5^{mm} alti latera 11^{mm} longa cartilaginea pallida reticulato-nervosa exlobata. Petala 23^{mm} longa rosea. Capsula calyce 3-plo superata, valvis acute triangularibus 3^{mm} longis sicut styli basis 4^{mm} longa cartilagineis pallidis, seminibus vix 1^{mm} longis.

Guatemala, Depart. Alta Verapaz: in fundo *Sepacuité* dicto, Mart. 1902, *Cook et Griggs* (n. 106), Apr. 1902, *Cook et Griggs* (n. 575); in declivibus humidis inter Senahú at Actalá, Jan. 1905, *Maxon et Hay* (n. 3331); secus viam inter Sepacuité et Secanquiń, alt. 1200^{m}, Maj. 1905, *H. Pittier* (n. 314).

Conostegia dolichostylis Donn. Sm.—Tota praeter foliorum paginam superiorem plus minus dense stellato- et ferrugineo-pubescens. Folia lanceolata superne subsensim tenuiterque, deorsum brevius, acuminata obsolete sinuato-denticulata 5-plinervia. Flores pauci graciliter pedicellati 5-meri inter maximos. Stylus stamina longe superans.

Ramuli teretes et petioli dense pubescentes. Folia membranacea 8–15^{cm} longa 3–5^{cm} lata leviter disparia supra glabra subtus nervis venisque pubescentia et demum inter nervos glabrescentia, nervo utroque superiore 10–22^{mm} supra basin limbi prodeunte, petiolis 1–3^{cm} longis. Panicula in exemplo unico manco suppetente 3-flora ut videtur, pedunculo 5^{mm} longo, pedicellis 11^{mm} longis. Calycis tubus turbinato-campanulatus 7^{mm} longus atque latus pubescens. Petala obovata 12^{mm} longa. Antherae crasso-oblongae 5.5^{mm} longae filamenta aequantes. Stylus 15^{mm} longus, stigmate hemispherico 1^{mm}-diametrali. Alabastra ignota.—*C. arboreae* Schauer proxima.

In silvis ad Buenos Aires, Comarca de Puntarenas, Costa Rica, Febr. 1892, *Tonduz* (n. 4943).

Conostegia rhodopetala Donn. Sm.—Folia glabra coriacea oblongo-lanceolata caudato-acuminata basi acuta integra triplinervia. Panicula pyramidata decomposita, axibus primariis 4-verticellatis. Calycis tubus campanulatus limbus conicus rostratus. Petala obovato-orbicularia.

Arbor, ramulis obtuse tetragonis, junioribus uti folia nascentia et panicula leviter furfuraceis. Folia decussata in eodem jugo satis inaequalia 15–20^{cm} longa 5–8^{cm} lata in caudam linearem 8–10^{mm} longam desinentia, nervis paulo supra basin egredientibus validis, duobus basalibus submarginalibus tenuibus interdum adjectis, venis transversis 4–6^{mm} inter se distantibus, petiolis 2–5^{cm} longis. Panicula 15–20^{cm} alta densiflora, pedicellis 7–12^{mm} longis, floribus 6-meris. Alabastra rostello 2–3^{mm} longo adjecto 9^{mm} longa 3^{mm} crassa. Petala 6^{mm} longa rosea. Stamina 12–14, antheris leviter arcuatis. Stylus 4^{mm} longus crassus, stigmate capitato.—Juxta methodum in Monographia clari COGNIAUX expositam apud *C. Pittieri* Cogn. collocari debet.

In sylvis ad La Palma, Prov. San José, Costa Rica, alt. 1500–1800^{m}, Jul. 1895, *Tonduz* (n. 9702), Sept. 1896, *Pittier* (n. 10169), Maj. 1898, *Tonduz* (n. 12347).—Las Lajas, San Isidro, Prov. Heredia, Costa Rica, alt. 1500^{m}, Sept. 1900, *Pittier* (n. 14022).

Conostegia vulcanicola Donn. Sm.—Stellulato-pubescens. Folia supra glabrescentia subtus nervis venisque pubescentia lanceolato-elliptica utrinque acuminata margine setulifera quintuplinervia, nervis superioribus a basi remotis. Alabastra inter minima oblongo-ovoidea medio constricta superne conica apiculata. Flores 5–6-meri.

Ramuli teretes. Folia nascentia utrinque ferrugineo-tomentulosa, provectiora supra pilosiuscula vel glabrescentia subcoriacea 8–14^{cm} longa 3–5.5^{cm} lata in eodem jugo parum aequalia apice curvilineo-acuminata, nervis superioribus 1.5–2.5^{cm} supra basin limbi a medio secendentibus, setulis marginalibus cito caducis, petiolis tomentulosis 1.5–2.5^{cm} longis. Panicula pyramidalis trichotoma pubescens, pedicellis 1–3^{mm} longis. Alabastra 5–6^{mm} longa 3^{mm} crassa exrostrata basi subtruncata pube punctulata vel glabrescentia. Petala obovata 5^{mm} longa crenulata flava. Stamina 10–12, antheris rectis 2^{mm} longis. Stylus 4^{mm} longus, stigmate non dilatato. Fructus urceolato-globularis 4^{mm} diametralis glabrescens in sicco plumbeus.—Ad *C. Cooperi* Cogn. quibusdam notis accedens foliorum nervatione recedit.

In silvis ad Achiote in monte vulcanico *Pods* dicto, Prov. Alajuela, Costa Rica, alt. 2200^{m}, Nov. 1896, *Tonduz* (nn. 10836, 10840).

Miconia astroplocama Donn. Sm. (§ TAMONEA Cogn.)—Pilis stellatis luteo-fuscis tomentulosa. Folia supra nitida subtus praesertim ad nervos pilosa lanceolato-oblonga apice longe acuminata

basi acutiuscula valde disparia 5-nervia. Flores pedicellati terni 5-meri. Calyx lobatus petalis dimidio longior. Filamenta glabra, antheris profundiuscule bilobatis.

Arbor, coma rotundata, ramulis teretibus et petiolis panicula calycibus dense tomentulosis. Folia subcoriacea integra supra glabra et in herbario nigricantia subtus inter nervos pilis e papilla ferruginea pluriradiatis conspersa, cujusque paris folio altero 18–33^{cm} longa 5–12^{cm} lato quam minus breviter petiolatum 2–3-plo majore, petiolis crassis 2–5.5^{cm} longis. Panicula pyramidalis foliis brevior multiflora, cymulis trifloris, pedicellis 2–4^{mm} longis. Calyx anguste campanularis 4.5^{mm} longus limbo haud dilatato 2^{mm} latus, lobis parvis rotundatis extus tuberculo punctatis. Petala quadrato-oblonga 3^{mm} longa. Antherae lineares inaequales 3–4^{mm} longae incurvae fere usque ad 0.5^{mm} supra basin affixae. Ovarium semiliberum 3-loculare, stylo 9^{mm} longo. Bacca globosa 5^{mm} diametralis stellato pilosa.

In silvis ad fundum *Tuis* vocatum, Prov. Cartago, Costa Rica, alt. 650^{m}, Nov. 1897, *Tonduz* (n. 11438).—In silvis ad Las Vueltas, Tucurrique, Costa Rica, alt. 700^{m}, Dec. 1898, *Tonduz* (n. 12872).

Miconia nutans Donn. Sm. (§ LACERARIA Naud.)—Leviter furfuracea. Folia supra glabra subtus praetermissis nervis glabrescentia obovato-oblonga cordato-acuminata in petiolum canaliculatum attenuata subobsolete sinuato-denticulata 5-plinervia, venis transversalibus crebris. Flores 5-meri. Calyx medio constrictus. Petala lineari-oblonga. Ovarium 5-loculare.

Ramuli obtuse tetragoni et petioli panicula foliorum subtus nervi furfuribus minutis glandularibus stellatis ferruginosi. Folia coriacea siccitate utrinque laete viridia subtus primum furfuracea demum inter nervos furfurium reliquis rubropunctulata satis disparia 15–25^{cm} longa 7–10^{cm} lata, nervo utroque superiore circiter 5^{mm} supra basin limbi a medio secedente, inferiore submarginali, venis transversalibus angulo recto patulis simplicibus 3–5^{mm} inter se distantibus subtus conspicuis, petiolis 2–3 ^{cm} longis. Paniculae singulae vel ternae pyramidales foliis superatae apice nutantes repetitus trichotomae, floribus sessilibus vel breviter pedicellatis, bracteolis oblongis cito caducis. Calyx pallide furfuraceus 3.5^{mm} longus, tubo subgloboso, limbo dilatato 2.5^{mm} lato usque ad medium in lobos ovatos disrupto. Petala 4^{mm} longa 1^{mm} lata utrinque truncata nervata. Antherae vix 3^{mm} longae. Ovarium fere totum adnatum, stylo 7–8^{mm} longo. Bacca depresso-globosa 3^{mm} alta 4.5^{mm} crassa 10-costata limbo 2^{mm} alto coronata.—*M. aureoidi* Cogn. proxima.

Comarca de Puntarenas, Costa Rica, alt. 1100^{m}, Febr. 1897, *Pittier*, ad ripas Río Coto (n. 11059), in paludosis secus Río Cañas Gordas (n. 11060).—In fundo *Cubilquitz* dicto, Depart. Alta Verapaz, Guatemala, alt. 350^{m}, Oct. 1903, *von Tuerckheim*, n. 8522 ex Pl. Guat. &c., quas ed. Donn. Sm. Sub *M. obovali* Naud. olim distributa.

Blakea anomala Donn. Sm.—Folia obovato-elliptica abrupte caudato-acuminata basi acuta quinquenervia breviter petiolata. Pedunculi solitarii filiformes. Bracteae exiguae triangulares. Calyx primum clausus deinde in lobos 4 tubum aequantes dirumpens.

Saprogena. Rami teretes, ramulis petiolisque dense paleaceo-furfuraceis. Folia chartacea supra glabra subtus nervis leviter furfuracea in eodem jugo satis inaequalia consimilia 5.5–10^{cm} longa 3–5.5^{cm} lata in caudam 1–1.5^{cm} longam desinentia, venis transversis 0.5–1^{mm} inter se distantibus, petiolis 5–8^{mm} longis. Pedunculi leviter furfuracei 3–6^{cm} longi, bracteis 1.5–2.5^{mm} longis inaequalibus basi connatis, alabastris turbinatis, floribus praetermisso calyce 6-meris. Calyx coloratus cum bracteis tenuissime furfuraceus, tubo campanulato 7^{mm} longo totidemque lato, lobis deltoideo-ovatis intus densius furfuraceis. Petala obovato-oblonga 14–15^{mm} longa. Antherae pendulae connatae ovales filamentis rubris 6^{mm} longis bis superatae, calcare acuto. Stylus 7^{mm} longus.—Ad *B. gracilem* Hemsl. habitu accedens ab omnibus congeneribus bracteis necnon calyce insigniter recedit.

In truncis putridis ad La Palma, Prov. San José, Costa Rica, alt. 1450–1550^{m}, Sept. 1896, *Pittier* (n. 10165), Sept. 1898, *Tonduz* (n. 12521).

Passiflora Salvadorensis Donn. Sm. (§ EUDECALOBA Mast.)—Folia subtus glauca ima basi cuneata paulo infra medium biloba, lobis divergentibus paulo longioribus quam latioribus rotundatis. Pedunculi petiolos subaequantes. Calyx basi patelliformis, segmentis linearibus pedunculo bis terve brevioribus. Coronae faucialis fila petalis aequilonga, corona basali 5-crenata. Gynophorum gracillimum. Filamenta sicut styli perelongata.

Scandens glabra, ramis angulatis. Folia circumscriptione quasi semiovata 8–9^{cm} longa 6.5–8.5^{cm} lata membranacea subtus inter nervos glandulis magnis ocellata trinervia, lobis et sinu vix lobulato nervo exeunte aristulatis, nervis lateralibus angulo semirecto divergentibus, petiolis eglandulosis 3–5^{cm} longis, stipulis filiformibus. Pedunculi 2–3-ni medio filiformi-bracteati, alabastris e basi truncata oblongis. Sepala 16^{mm} longa. Petala hyalina linearia 8^{mm} longa. Corona faucialis filamentosa, filis uniseriatis linearibus purpureis. Corona mediana late tubulosa 3–4^{mm} alta plicata denticulata purpurea. Corona basalis 1–2^{mm} alta herbacea. Gynophorum 12^{mm} longum. Filamenta stylos subaequantia 8–9^{mm} longa. Ovarium globoso-ovale 1^{mm} longum. Fructus ignotus. —*P. ornithourae* Mast. affinis. Ab incolis *Calzoncillos* facete dicitur.

San Salvador, Republ. del Salvador, Maj. 1905, *Luis V. Velasco*, n. 8887 ex Pl. Guat. &c. quas ed. Donn. Sm.

Dendropanax querceti Donn. Sm.—Folia breviter petiolata lanceolata apice curvilineo-acuminata infra medium sensim angustata

calloso-denticulata. Umbella solitaria, pedunculo petiolis paulo longiore pedicellos vix superante ima basi bracteoso apice in receptaculum ferrugineo-bracteolosum dilatato.

Glabrum dichotomo-ramosum. Folia coriacea 5–10^{cm} longa 3–4-plo longiora quam latiora calloso-apiculata, nervis lateralibus utrinque 11–14, duobus infimis submarginalibus longius ascendentibus, reticulis minutis, petiolis 4–10^{mm} longis. Pedunculus robustus 6–12^{mm} longus, bracteis imbricatis scariosis ovatis mucronatis, receptaculi bracteolis paleaeformibus lanceolatis pulverulentis, umbella unica simplice 12–18-flora, pedicellis 5–10^{mm} longis. Calycis tubus obconicus 2^{mm} longus, margo mucronulis denticulatus. Petala ovata 1-nervia apice inflexa calycem aequantia. Stamina petalis aequilonga, antheris ovalibus. Discus a calyce angustissime liber. Styli in columnam connati. Fructus globosus 4–5^{mm}-diametralis.

In quercetis ad El Copey, Prov. Cartago, Costa Rica, alt. 1800^{m}, Mart. 1896, Mart. 1898, *Ad. Tonduz* (nn. 11826, 12196).

Rondeletia aetheocalymma Donn. Sm. (§ ARACHNOTHRYX Benth. et Hook.)—Glabra. Folia obovato-elliptica subabrupte acuminata basi acuta. Stipulae lineari-oblongae mucronulatae. Corymbus obpyramidalis trichotomo-cymosus, floribus pilosis. Calycis lobi elliptici, altero ceteris inaequalibus 2–3-plo majore quam tubus corollinus cylindricus bis breviore.

Frutex 3–4-metralis, ramis et foliis nitidis. Haec coriacea 12–15^{cm} longa 5–7^{cm} lata, nervis lateralibus utrinque 9–11, venulis subimmersis, petiolis 9–12^{mm} longis, stipulis 6–9^{mm} longis. Corymbus terminalis folia superans, pedunculo 4–11^{cm} longo, axibus semierectis pubescentibus, primariis foliaceo-bracteatis 3·5–5·5^{cm} longis, secundariis 1·5–2·5^{cm} longis, bracteolis linearibus 6–12^{mm} longis glabris, floribus tetrameris, dichotomali sessili, lateralibus brevissime pedicellatis. Calycis tubus cano-sericeus, lobi glabrescentes, maximo 7^{mm} longo 3^{mm} lato. Corollae albae tubus 13^{mm} longus cano-pilosus, os nudum exannulatum, lobi obovato-rotundi undulati intus farinosi. Capsula depresso globosa 6^{mm}-diametralis pilosa polysperma.—*R. linguiformi* Hemsl. proxima.

Secus viam inter Sepacuité et Secanquiń, Depart. Alta Verapaz, Guatemala, alt. 550–900^{m}, Jan. 1905, *Maxon et Hay* (n. 3275).

Rondeletia stachyoidea Donn. Sm. (§ ARACHNOTHRYX Benth. et Hook.)—Folia anguste lanceolata utrinque, praesertim sursum, attenuata supra parce pilosa subtus strigillosa. Thyrsus spiciformis, capitellis subsessilibus 2–3-floris. Calycis arachnoideo-pilosi lobi lineari-lanceolati paulo inaequales. Corolla praeter lobos extus basi barbatos glabra. Capsula parva.

Arbuscula quinquemetralis, ramulis appresse pilosis, internodiis, saltem superioribus, brevissimis. Folia 10–16^{cm} longa 2–3.5^{cm} lata sursum tenuissime acutissimeque attenuata supra cito glabrescentia, nervis lateralibus utrinsecus circiter 9 longe ascendentibus subtus conspicuis, petiolis 5–10^{mm} longis pilosis, stipulis lineari-lanceolatis filiforme attenuatis 1.5^{cm} longis internodia superiora aequantibus. Thyrsus terminalis 9–11^{cm} longus pilosus, bracteolis lineari-lanceolatis calycem aequantibus arachnoideo-pilosis, floribus subsessilibus tetrameris. Calycis lobi 3–4^{mm} longi. Corollae albae tubus tenuis 8^{mm} longus sursum leviter ampliatus, os nudum exannulatum, lobi rotundati. Capsula globosa 2^{mm}-diametralis pilosiuscula polysperma.—*R. gracili* Hemsl. affinis differt inter alia capitellis paene sessilibus paucifloris.

Semococh, Depart. Alta Verapaz, Guatemala, alt. 600–900^{m}, Apr. 1905 (*Robert Hay*).

Rondeletia Thiemei Donn. Sm. (§ ARACHNOTHRYX Benth. et Hook.)—Pilosa. Folia parva lanceolato-elliptica utrinque, sursum longius acutiusque, curvilineo-attenuata subsessilia supra scabriuscula subtus cinereo-pilosa. Panicula sessilis parva ovalis, axibus racemiformibus vel cymuliferis. Calycis segmentum quartum lanceolatum ceteris subulatis 3-plo longius.

Frutex, ramulis et panicula cinereo-velutinis. Folia 5–8^{cm} longa 2–2.5^{cm} lata supra punctulata, stipulis triangularibus subulato-acuminatis 3–4^{mm} longis. Panicula terminalis 5–6^{cm} longa foliaceo-bracteosa, axibus patentibus, bracteolis linearibus 4–6^{mm} longis, pedicellis 1–3^{mm} longis, floribus tetrameris singulis vel 2–3-nis, cymulis pedicellatis. Calycis pilosi segmentum maximum 5–6^{mm} longum 3-nervium. Corollae tubus pilosus tenuiter cylindricus 8–10^{mm} longus, os nudum exannulatum, lobi subrotundi 3^{mm} longi crenati utrinque glabri. Capsula globosa 5^{mm}-diametralis pilosa lobis calycinis coronata, seminibus numerosis oblongo-cubicis scrobiculosis.

Río Chamelecón, Depart, Santa Bárbara, Honduras, alt. 400^{m}, Jan. 1888, *C. Thieme*, n. 5276 ex Pl. Guat. &c. quas ed. Donn. Sm.

BIDENS COREOPSIDIS DC., var. **procumbens** Donn. Sm.—Plerumque 5-foliolata, foliolis tenuiter lanceolatis subglabris, corymborum axibus filiformibus, acheniis margine dense barbatis, aristis parce hispidulis.

Prope Secanquiḿ, Depart. Alta Verapaz, Guatemala, alt. 550^{m}, Jan. 1905, *Maxon et Hay* (n. 3162).

Hectia dichroantha Donn. Sm.—Paniculae rami abbreviati simplices continue denseque racemoso-floriferi, racemis cylindricis, floribus singulis. Perianthii segmenta acuta subaequalia pedicello bis breviore adjecto bracteolam paulo superantia.

Folia 3.5–4^{dm} longa perlonge attenuata supra glabra subtus niveo-lepidota spinis corneis inter se 5–10^{mm} distantibus 1–3^{mm} longis crenulata. Scapus cum panicula glaber in sicco purpurascens, vaginis supra oinochrois subtus niveis e basi ovato-lanceolata longissime filiformeque attenuatis 7–9^{cm} longis internodia superantibus. Panicula 5–6^{dm} longa, racemis raro binis sueto inter se 3–7^{cm} distantibus 4–8^{cm} longis, bracteis 1.5–2^{cm} longis uti bracteolae 7–8^{mm} longae lineari-lanceolatis, pedicellis 3^{mm} longis, floribus glabris. Perianthii segmenta discreta ovato-lanceolata, exteriora 5^{mm} longa cum bracteis bracteolisque oinochroa, interiora 6^{mm} longa alba. Florum masculinorum solum cognitorum stamina perianthium aequantia, ovarii rudimentum glabrum.

In collibus declivibus ad flumen *Quililá* prope Santa Rosa, Depart. Baja Verapaz, Guatemala, alt. 1600^{m}, Maj. 1905, *O. F. Cook*.

BALTIMORE, MD.

A BACTERIAL DISEASE OF OLEANDER.

BACILLUS OLEAE (Arcang.) Trev.

CLAYTON O. SMITH.

(WITH FOUR FIGURES)

DURING the autumn of 1905 some diseased oleanders were sent from a nursery to the plant pathological laboratory of the University of California. This disease has been occasionally reported as occur-

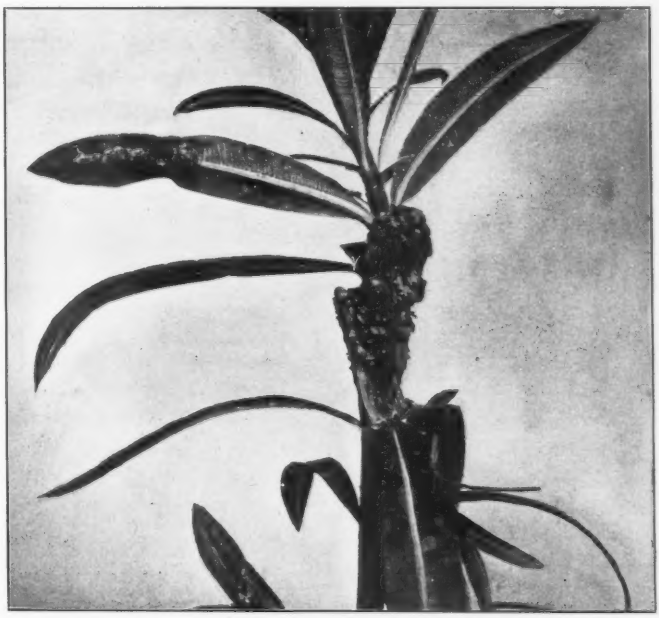


FIG. 1.—Oleander, showing knots on stem and leaf from natural infection.

ring on young oleanders in this state. The trouble affects the stem and leaves (*figs. 1, 2*), forming large, hard, woody knots. These knots were examined by Professor R. E. SMITH and found to contain

numerous bacteria which were produced in small colonies in the tissue. The general nature of the knot and the close botanical relationship of the oleander and olive immediately suggested to him that the trouble might be caused by the same organism that produces the so-called knot or tuberculosis of the olive. The subject was assigned to the writer for further investigation. The work for the most part was done at the bacteriological laboratory of the University, and thanks are due to C. M. HARING and Professor A. R. WARD, of that laboratory, for their courtesy and suggestions during the investigation.

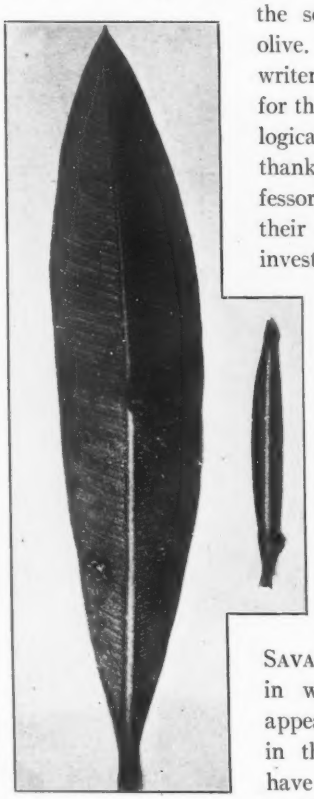


FIG. 2.—Leaves of oleander, showing knots from natural infection.

The olive knot is a disease of the branches and leaves of the olive tree. It occurs in Egypt, in the olive-growing sections of Europe bordering on the Mediterranean, and is also found in California where the olive is grown. The disease has been known for many years and is even described by Roman writers; but its bacterial origin has only been recognized since 1886, when the organism was discovered by ARCANGELI (1) and SAVASTANO (2). It is found in the knots in what may be called colonies. These appear as clearer or more transparent spots in the callus-like tissue. These growths have their origin near the cambium layer and at length become darker in color. About this colony hypertrophy of the tissue takes place, as a natural effort of the plant to heal the injury caused by the bacteria. This same process of healing would take place in mechanical injuries. The result is that much soft, spongy tissue is formed that makes a rather favorable place for new bacterial growth, which means a new

formation of callus tissue and hence an increase in the size of the knot.

These large knot-like growths of the olive may be found on the leaves, branches, and trunks, few in number or in great abundance. They must not be confused with those caused by insects, and they are also distinct from the enlargements formed on the roots of the Leguminosae by bacteria. Badly diseased trees show scant foliage, limited growth, and occasional dead branches.

The cultural characteristics of the olive knot organism have been studied by SAVASTANO (**5**) and seem to agree quite well with those observed during this study. The following is a portion of the translation of SAVASTANO'S account of the disease as published by PIERCE (**7**):

> This microorganism is a bacillus of medium size; length three to four times its width; it is isolated, but is sometimes joined into chains; the extremities are slightly rounded off. In drops of bouillon it has a distinct movement. The colony has a variable form, from round to oval, with a well-defined margin. In the beginning it is uniformly pointed; later it forms one or two peripheral circles. It is whitish by reflected light, cedar color by transmitted light. The bacillus lives well in ordinary culture media (bouillon, potato, gelatin, agar). The culture has a relatively long life; cultures made in March were still living in June. In short, degeneration begins in about three months. On potato it lives very well and develops rapidly; the colonies are at first like so many round dots, translucent straw color, which, as they develop, form on the surface of the potato a uniform stratum, translucent, and of a deeper color. The bacillus acquires greater dimensions. On gelatin plates it lives very well, with characters and forms as above indicated. In the tubes of gelatin (*a becco*) the culture presents the appearance of a uniform stratum, whitish, the margin finely bilobed, reminding one of the margin of a leaf, the whole culture taking the form of a spatulate leaf. It is slightly dichroic. In tubes of agar (*a becco*) the culture is identical with the preceding, the margin is less bilobed. The culture by needle in gelatin presents a uniform, transparent, finely pointed appearance. On the surface of the meniscus the form is irregularly rounded, with a finely lobed margin, as in the preceding.

In the study of the present disease, the organism was first isolated from the oleander in the manner described below. Inoculations were then made on the oleander and olive. Positive results were obtained in all cases. No effort was made to try inoculations on other plants. SAVASTANO (**5**), however, failed to make successful inoculation with the olive knot organisms on peach, plum, apricot,

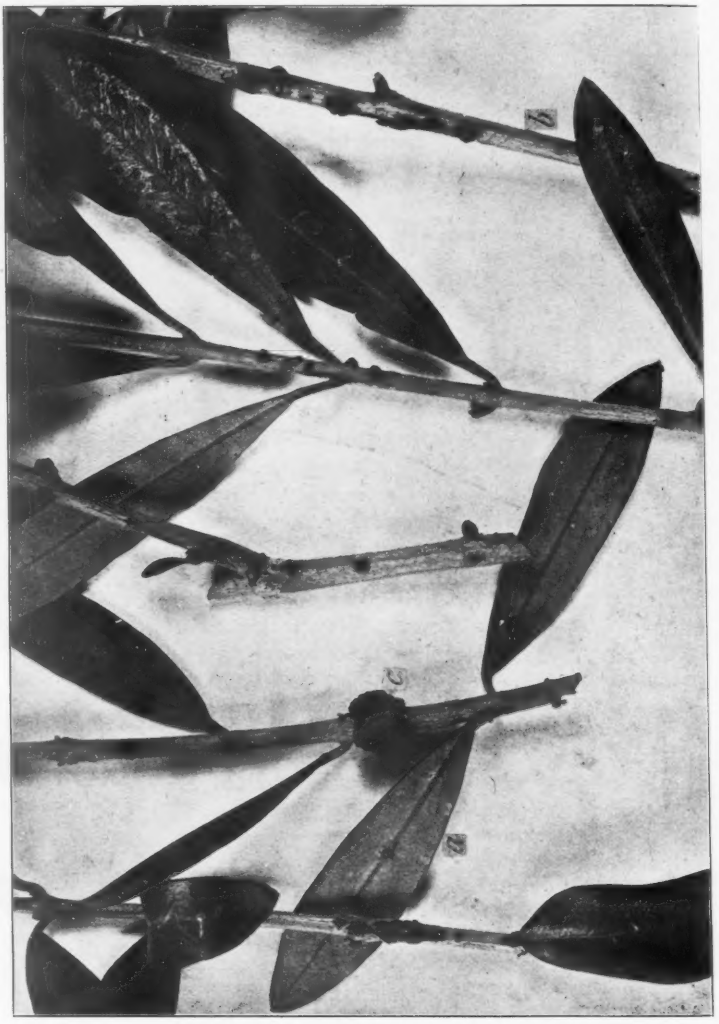


FIG. 3.—Knots on olive, caused by artificial infection from pure culture: *a*, on leaves; *b*, on tree inoculated in the open; *c*, large knot and smaller knots on plants growing under glass.

grape, fig, pear, apple, bitter orange, lemon, rose, *Abies excelsa*, *A. pectinata*, and *Cedrus Libani*. He did not experiment with the oleander, however, or any other plant related to the olive.

It required considerable time in the writer's experiments for the disease to develop, though in a month's time the first indication of tissue enlargement could be observed. This continued to increase until on the olive quite a knot was formed in two month's time (*fig. 3*). Often the infection did not produce such a large knot as indicated in *fig. 3*, but smaller swellings of the tissue. In this inoculation work agar cultures, 48 hours old, were used, except on one occasion when a bouillon culture was tried. Either gave equally satisfactory results. The visible effects of the inoculations showed sooner on the oleander, but their size and the rapidity of knot-formation seems to depend upon the rapidity with which the plant is growing, as has been before observed by SAVASTANO (**4**) in his study of the olive knot. The organism grows on both the stem and the leaves. In one experiment a leaf of an oleander near the top of the plant was inoculated on the midvein; this inoculation grew well, and from it secondary natural infections resulted on the stem. It was not difficult to trace the new infections on the stem to the very base of the petiole of the diseased leaf. Infection (*fig. 4*) took place probably through the stomata and lenticels. Checks were used by making punctures with a sterile needle, but these gave no knot formations.

The lesions and growth on the hosts were quite different. On the oleander at first there was a slight enlargement of the tissue that became somewhat rounded at the point of infection. After a time, as the new growth continued, there was a splitting of the epidermis in a longitudinal direction, forming a cleft (*fig. 4*). After this a spongy growth formed, which is rather dark in color and contains numerous small colonies of the bacteria. On the olive there was the same enlargement of the tissue as in the oleander, but the formation of the new growth was much more rapid, regular knots being soon formed by the growing out of the new tissue. This took place in the olive rapidly, while in the oleander it was only in the advanced stages that this new callus tissue grew into a knot-like formation. The knots on the olive agreed in appearance with specimens of the typical knots as it occurs in California, and with the illustration as given by PIERCE (**7**), BIOLETTI (**8**), and ERWIN SMITH (**9**).

The original cultures were made from the diseased tissue of the oleander by first cutting away the outside tissue with a scapel, steri-

open; *c*, large knot and smaller knots on plants growing under glass.

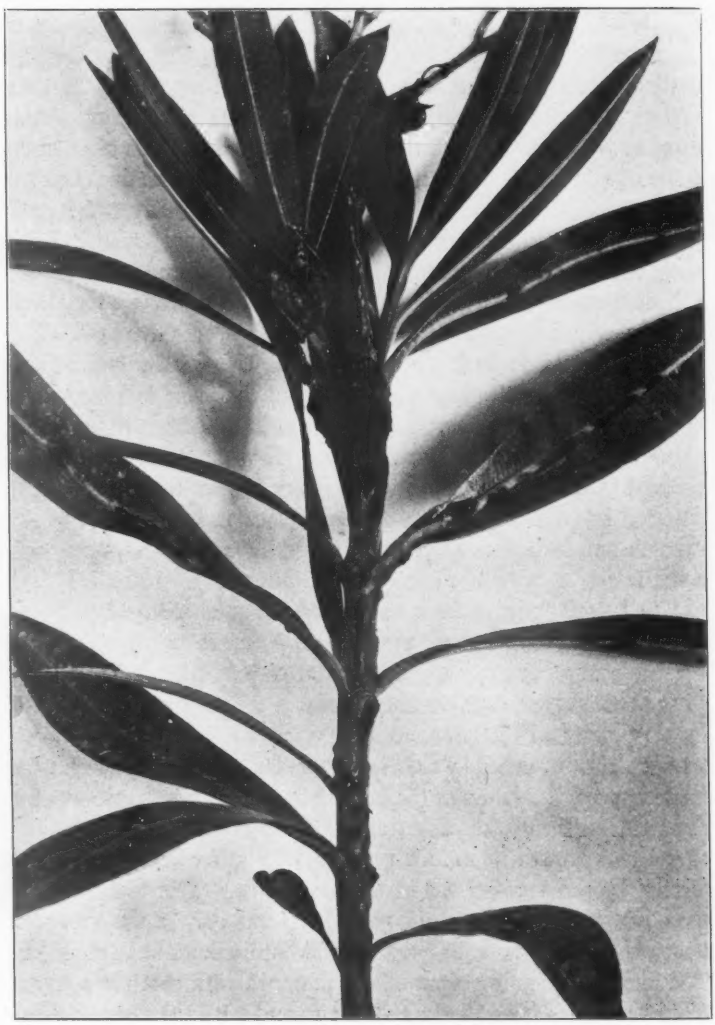


FIG. 4.—Knots on oleander leaves and stem caused by artificial infection from pure culture.

lized by flaming. When this was done, the small colonies could be seen in the tissue as dark clear places; these were touched with a sterile platinum needle, and a tube of ordinary meat bouillon was inoculated. Several such tubes were inoculated from the knots, and in about two or three days there was abundant growth. From these tubes dilution cultures were made on agar in Petri dishes. Other dilution cultures in Petri dishes were then made from ten of the bouillon tubes, and the uniformity with which one form of colony appeared in the Petri plates seemed to indicate quite conclusively that this was the organism causing the disease. Transfers were then made from these colonies to agar tubes and thence to various culture media.

CULTURAL CHARACTERISTICS.—The organism was grown on the ordinary culture titrated to +1.5 to phenolphthalin and grown at room temperature.

Morphology.—The organism is a motile rod with rounded ends, 1.5–2.5×0.5–0.6 μ. It is usually solitary, but may occur in pairs. Organism direct from the plant as well as from the pure culture shows motility. The size as above given was from a preparation made directly from the tissue.

Agar slant.—On agar growth appeared in about twenty-four hours, as a very thin, grayish-white surface growth. This spread quite rapidly over the surface, especially near the lower portion of the tube. Sometimes small roundish colonies appeared at the side of the growth along the stroke. In cultures a week old the growth is white by transmitted and reflected light. The growth is very thin and scarcely perceptible at first. The condensation water becomes clouded and white.

Agar plate colonies.—On agar plates the colonies become visible after three days. The deeper ones are small, globose, or biconvex with sharp entire margins. The surface colonies are larger and more spreading, circular in outline, whitish in color, and somewhat more dense in center than at margin. The deeper ones often have a straw or cedar color as observed by SAVASTANO. The surface colonies measure 2–4^{mm} in diameter when four days old. They are more vigorous than colonies in gelatin of the same age.

Glycerin agar.—Growth much the same as in agar, except more

vigorous, and hence a thicker surface growth resulted. Numerous small colonies also developed on surface.

Gelatin stab.—Growth takes place along the stab and also on the surface of the media. The growth along the puncture was filiform; on the surface it was thin, spreading from the point of puncture, the margin undulatory to lobate-lobular. No liquefaction.

Gelatin plate colonies.—These appeared in two to three days. The deep colonies much the same as those in agar Petri plates. Surface colonies more spreading, forming a very thin growth with irregular undulatory margins, some darker in color than surface colonies. Surface colonies under low power showed the center to be denser than the margin and finely reticulated. Toward the margin the reticulations are much coarser than in the center.

Potato.—Growth was vigorous and characterized by always being straw color. The growth was quite markedly raised above the surface and soon covered the entire plug.

Bouillon.—In meat bouillon growth appeared after two days as a fine granular substance that remained in suspension. The culture is at first slightly acid to litmus, but becomes alkaline after two weeks.

Glucose bouillon.—Growth the same as in the meat boullion, with the same reactions. The change from acid to alkaline reaction is much slower than in the other media.

Saccharose bouillon.—Growth at first more vigorous than in either the bouillon or glucose bouillon; acid at first, neutral after seven days, and alkaline in two weeks.

Lactose bouillon.—Showed the same general characteristics as saccharose, acid at first, then neutral and alkaline after two weeks.

Litmus milk.—Showed no change until ten days, when there was a distinct alkaline reaction. No coagulation ever occurred. After fifteen days the medium was quite blue, with a slight whitish precipitate at the bottom of the tube. This was not granular, but subgelatinous, and when shaken into solution settled again to the bottom. In a tube two months old the liquid becomes very blue and alkaline.

Milk.—Showed no change except a slight yellowing in color.

Scarcely any experimental work was done in growing the Bacillus at different temperatures. SAVASTANO (**4**) states the optimum temperature for the olive knot to be between 32–38° C, and SMITH (**9**)

has still further restricted the limits of best growth to between 35–37.8° C. Several experiments were conducted by the writer with the oleander organism in temperature about 35.5° C, and no growth took place on agar, bouillon, or potato after four days, although there was good growth in the inoculated bouillon tube at room temperature.

All the liquid media become at first acid to litmus, but change to an alkaline reaction in about two weeks. The media used were all titrated to +1.5 to phenolphthalein. This would be slightly alkaline to litmus, so the growth of the organism caused first an acid, then an alkaline reaction. All the cultural characteristics of the oleander organism, so far as possible, were compared with those described from the olive organism. There seems to be a very close agreement, and without question the organism is identical.

From knots produced by artificial inoculations on olive and oleander the organism was isolated (in the same manner as described before) and grown in the same media as was the original culture from the oleander. These two series agreed perfectly in culture characteristics with one another and with the original culture.

The organism also was isolated from naturally infected olive knots in the same manner as for the oleander. Growth on various culture media showed biochemical and cultural characteristics that agreed with those observed by the writer for the oleander knot; and with those described by SAVASTANO in his study of the olive knot.

This oleander disease is not believed to be a new trouble, but similar to the one found on olives. They are both caused by a motile rod (Bacillus) that grows well on the ordinary culture media, and will cause infection of the olive and the oleander. This infection at length causes characteristic lesions and knot-like growths on the stem and leaves. The knots produced on the olive by the oleander organism agree with typical knots as found on cultivated olives in California, and with various illustrations of the olive knot. The cultural characteristics of the two are similar in all essential respects.

LABORATORY OF PLANT PATHOLOGY,
University of California, Berkeley.

LITERATURE CITED.

1. ARCANGELI, G., Sopra la malattie dell' olivo detta volgarmente rogna. Pisa. 1886.
2. SAVASTANO, L., Les maladies de l'olivier et la tuberculose en particulier. Compt. Rend. **103**: 1144. 1886.
3. ———, Les maladies de l'olivier; hyperplasies et tumeurs. *Idem* 1278. 1886.
4. ———, Tuberculosi, perplasie e tumori dell' olivo. I e II Memoria. Naples. 1887.
5. ———, Il baccillo della tuberculosi dell' olivo, nota suppletiva. Rend. Lincei **5**: 92–94. 1889.
6. PRILLIEUX, E., Les tumeurs à bacilles de l'olivier, etc. Compt. Rend. **108**: 249. 1891.
7. PIERCE, NEWTON B., Tuberculosis of the olive. Jour. Mycol. **6**: 148–153. 1891.
8. BIOLETTI, F. T., The olive knot. Calif. Agric. Exp. Sta. Bull. 120. 1898.
9. SMITH, ERWIN F., Bacteria in relation to plant diseases. **1**: 10. 1905.

CURRENT LITERATURE.

BOOK REVIEWS.

Another botanical dictionary.

THIS Italian book is in a measure both a dictionary, with its mere definition of terms, and an encyclopedia, with its more elaborate treatment of topics and brief biographies of celebrated botanists.[1] In its plan, therefore, it departs widely from JACKSON'S *Glossary;* and it departs from it still more widely in mechanical execution. For it is published by ULRICO HOEPLI of Milan whose series of dictionaries and manuals is famous. It is 11×16^{cm}, printed on thin flexible paper, with narrow margins and small but clear type, bound in half vellum, and though it contains nearly a thousand pages it is less than 4^{cm} thick. Every detail is appropriate to its use and the book is therefore a model of convenience.

In substance the work is unexpectedly good. The definitions of the simpler terms are usually clear and succinct, and the treatment of topics is full enough and is accompanied by citation of enough of the important literature to make the book of real value for reference. A few figures will indicate the extent of some topics. Thus to *nutrizione* are devoted nearly 36 pages; to *tessuti*, 10 pages; to *simmetria*, 6 pages; to *variazione*, 7 pages; to *germogliamento*, 6 pages; to *classificazione*, 8 pages; and so on. On the whole the assignment of space is made with good judgment.

The only adverse criticism that need be made is that the author has apparently not always assimilated fully the modern morphology that he is endeavoring to state. In consequence seeming contradictions and limitations occasionally appear. Thus the *sporofito* seems to belong only to ferns and spermatophytes; while the *sporogonio* appertains to the bryophytes. *Sporangio* is defined and discussed only with reference to pteridophytes, and *spore* would seem to be restricted to "cryptogams." Yet in other topics the relations of the microspores of spermatophytes are recognized and properly described, while the alternation of generations and heterospory are concisely but clearly treated.

On the whole there are probably as few shortcomings as could be expected and as many excellences as could be attained by any one who undertakes such a task. Cooperation, however, is fast coming to be necessary in work of such a scope.—C. R. B.

---

[1] BILANCIONI, EUG., Dizionario di botanica generale. Milano: Ulrico Hoepli. 16mo. pp. xx+926. *L* 10.

MINOR NOTICES.

Botanical papers at the St. Louis Congress.—Volume V[2] of the series being published by the Congress of Arts and Science held in connection with the St. Louis Exposition contains the botanical papers. The department of biology was divided into eleven sections, and twenty-five principal papers were read. Those of special interest to botanists, in the order of their appearance in the volume, are as follows: Development of morphological conceptions, JOHN M. COULTER; The recent development of biology, JACQUES LOEB; A comparison between artificial and natural selection, HUGO DEVRIES; The problem of the origin of species, C. O. WHITMAN; Plant morphology, F. O. BOWER; The fundamental problems of present-day plant morphology, K. GOEBEL; The development of plant physiology under the influence of the other sciences, J. WIESNER; Plant physiology, present problems, B. M. DUGGAR; The history and scope of plant pathology, J. C. ARTHUR; Vegetable pathology an economic science, M. B. WAITE; The position of ecology in modern science, O. DRUDE; The problems of ecology, B. L. ROBERTSON; Relations of bacteriology to other sciences, E. O. JORDAN; Some problems in the life history of pathogenic micro-organisms, THEOBALD SMITH.

All of these papers have been published in various journals, notably in *Science*, but it is convenient to know that they are all accessible in a single volume.—J. M. C.

Index Filicum.—The eleventh fascicle of CHRISTENSEN'S work has just appeared,[3] carrying the references from *Trichomanes Giesenhagenii* to the end of the list. There follow a list of additions, a list of corrections, and the beginning of a catalogue of literature arranged alphabetically.—J. M. C.

Trees of Java.—KOORDERS and VALETON[4] have published another fascicle of additions to the known arboreal flora of Java, containing the Moraceae. Seven genera are represented, including 95 species, 83 of which belong to Ficus, under which two new species are described.—J. M. C.

Cyperaceae.—The second part of HUSNOT'S synopsis of the Cyperaceae of France, Switzerland, and Belgium has appeared,[5] completing the list of species, and closing with a full index.—J. M. C.

---

[2] Congress of Arts and Science, Universal Exposition, St. Louis, 1904. Edited by HOWARD J. ROGERS. Volume V. Biology, Anthropology, Psychology, Sociology. pp. xi + 882. Boston and New York: Houghton, Mifflin and Company. 1906.

[3] CHRISTENSEN, C., Index Filicum, etc. Fasc., 11. Copenhagen. H. Hagerup. 1906. 3*s*. 6*d*.

[4] KOORDERS, S. H., and VALETON, TH., Boomsoorten op Java. Bijdrage no. 11. Mededeel. Depart. Landb. no. 2. pp. 277. Batavia, 1906.

[5] HUSNOT, T., Cypéracées: descriptions et figures des Cypéracées de France, Suisse et Belgique. Part II. pp. 49–83. *pls. 13–24*. Cahan, par Athis (Orne): the author. 1906. 5*fr*.

NOTES FOR STUDENTS.

Mosaic disease of tobacco.—In an extensive account of the mosaic disease of tobacco, which he has been investigating for a number of years, HUNGER[6] deals somewhat radically with the theories that have been advanced to account for the disease, and gives, as he believes, a new explanation. The earlier bacterial theories of MAYER, PRILLIEUX, DELACROIX, and others are treated only as matters of historical interest, since they are based on insufficient evidence. The more recent work of IWANOWSKI receives a more extended notice, although his view of the bacterial nature of the disease is likewise refuted, as HUNGER has been unable in his own investigations to corroborate IWANOWSKI'S work. BEIJERINCK'S theory that the disease is caused by an active fluid substance, itself capable of growth, is discredited on the ground that BEIJERINCK was unable to show that the virus was able to increase in quantity outside of the plant, and that his proof of the fluid nature of the virus (diffusion in agar) is not sufficient. Against the enzyme theory of WOODS the author raises two principal objections: (1) the transferability of the disease without limit does not accord with the properties of enzymes, whose activity is diminished by extreme dilution; (2) the virus of the mosaic disease has the property of being able to diffuse through parchment, a property not possessed by enzymes.

HUNGER advances the view that the mosaic disease is due exclusively to disturbances of metabolism, the outward manifestation of which is the peculiar form of variegation seen in the leaves. That the mosaic disease, to whatever cause it may be attributed, is a result of disturbances of the metabolic processes of the plant is beyond cavil; how this statement brings us any nearer to an explanation of the ultimate cause of the disease is beyond our comprehension. The author regards the disease as a sort of latent property possessed by tobacco plants, in which it may develop spontaneously if conditions are favorable, or to which it may be communicated by grafting and other methods. It is to be regarded as a kind of communicable variability! The active cause of the disease he regards as a toxin normally produced in the plant, but not injurious except under special conditions, when it accumulates in excess of the normal amount. The toxin is not like BEIJERINCK'S substance, capable of active growth, but is capable, when entering into a normal cell, of producing there catalytic effects, in consequence of which the toxin is there regenerated secondarily. In the words of the author, it is physiologically autocatalytic, all of which is perhaps merely a more extended theoretical explanation of what is ordinarily termed growth. HUNGER discusses also the etiology of the disease, and the probable relation of the methods of selection of tobacco practised at Deli to the rapid increase of the disease in the Sumatra tobacco districts. He points out that in order to obtain a high grade wrapper-leaf it has for generations been the practice of the tobacco growers to select for seed the plants with the thinnest leaves. This

[6] HUNGER, F. W. T., Untersuchungen und Betrachtungen über die Mosaik-Krankheit der Tabakspflanze. Zeitschr. Pflanzenkr. **15**:257–311. 1905.

selection has resulted in a race of plants degenerate from the standpoint of their power to resist unfavorable conditions. In the fields, even under usual conditions, a large percentage of the plants wilt on hot days. The reduction of thickness of the leaf is held in large measure responsible for the occurrence of the mosaic disease. A lesser though important influence is also attributed to the direct action of the soils.—H. HASSELBRING.

Microsporangia of Pteridosperms.—In 1905 KIDSTON published a preliminary note on the occurrence of microsporangia in connection with the foliage of Lyginodendron.[7] He has now published the full paper,[8] with detailed discussion and illustration. He first elaborates the evidence that *Sphenopteris Höninghausi* Brongn. and *Lyginodendron Oldhamium* Williamson are identical plants, and of course *Crossotheca Höninghausi* is the fertile pinnule of the former. It follows that the sporangia found on this species of Crossotheca are the microsporangia of *Lyginodendron Oldhamium*, a well-known pteridosperm. This rules out Miss BENSON'S claim that *Telangium Scotti* represents the microsporangia of this Lyginodendron.

The microsporangia are borne on modified pinnae, associated with sterile pinnae. The fertile pinna is oval, entire, on a short thick pedicel, and on the under side of the blade six to eight fusiform bilocular sporangia are borne, which bend inward at an early stage so that their pointed apices meet, forming a sort of hemispherical sorus. Later they spread apart and appear as a fringe hanging from the margin of the pinnule. In all cases the microspores are present, but no internal structure seems to be evident. The genus Crossotheca contains eight species, a new one being described in this paper, and perhaps it is safe to assume that all of them belong to the pteridosperms.

In a general discussion of the occurrence of fern-like plants, KIDSTON comes to the conclusion that the pteridosperms (including all Cycadofilices) are undoubtedly the oldest group of fern-like plants of which we have evidence, being plainly represented in the upper Devonian; that in the Lower Carboniferous pteridosperms were still dominant, true ferns being feebly represented, if at all, by the Botryopterideae; that in the Upper Carboniferous the same relative representation continued. It seems highly improbable, therefore, that the pteridosperms could have descended from true ferns, and KIDSTON is inclined to believe that there is no more relationship between the two groups than that of a common ancestry for pteridosperms and eusporangiate ferns. In summing up his conception of the most probable lines of descent of the existing ferns and cycads, he indicates three lines: an independent one, leading from Botryopterideae (of unknown origin) to the leptosporangiate ferns, and two lines converging in common but unknown ancestral forms, one leading to the Marattiaceae, the other through pteridosperms to the cycads.—J. M. C.

[7] BOT. GAZETTE **41**:219. 1906.

[8] KIDSTON, ROBERT, On the microsporangia of the Pteridospermae, with remarks on their relationships to existing groups. Phil. Trans. Roy. Soc. London B. **198**:413–445. *pls. 25–28.* 1906.

Experiments with Hieracium.—OSTENFELD and ROSENBERG have undertaken a series of experimental and cytological studies of the species of Hieracium. ROSENBERG has published[9] a preliminary report of some of his results; but the first paper of the series has just been received. It is by OSTENFELD,[10] reporting the results of castration and hybridization experiments. The castration experiments, carried on in 1903, 1904, and 1905, resulted in showing that in the genus Hieracium there are apogamous species, non-apogamous species, and transition species, and that the three subgenera conform in a general way to this division; Stenotheca having typical fertilization (the most primitive stage); Pilosella being intermediate, with both apogamous and non-apogamous species (the former in the majority); and Archieracium being entirely apogamous (except the *H. umbellatum* group). Attention is called to the fact that Taraxacum has "gone a little farther," all its species being apogamous.

The hybridization experiments are only in their inception, but the following results may be noted: a hybrid was produced between *H. pilosella* and *H. aurantiacum*, with greatly reduced fruiting power; *H. excellens*, itself probably a natural hybrid and producing only abortive pollen, gave hybrids by crossing with *H. aurantiacum* and *H. pilosella;* the hybrids arising from the same cross are heterogeneous; the fruiting power of hybrids is very slight.—J. M. C.

Infection experiments with mildews.—REED[11] has added a further contribution to the work inaugurated by NEGER and extended by MARCHAL and SALMON, bearing on the transferability of physiological forms of the Erysipheae from one specific host to another within a closely related group of plants. The results in general confirm the conclusions of previous investigators; namely, that there exist races of *E. graminis* and other species of mildews which have become specifically adapted to a single species, or more rarely to several species of one genus of host plants. REED finds, for example, that E. *graminis* from *Poa pratensis* will not infect other species of Poa except in some instances. Thus *P. nemoralis* is sometimes infected, while *P. trivialis* and *P. compressa* are infected very rarely. By this and other investigations the fact that a high degree of specialization exists in the Erysipheae, as in other groups of fungi, has been definitely established. While further demonstration of the existence of biological races is of less importance, the facts ascertained in this field of research furnish an excellent basis for other investigations, as for instance, the question of the permanency of these races, the problems connected with abnormal predisposition of the host plants, and others, some of which have already received attention. —H. HASSELBRING.

---

[9] Ber. Deutsch. Bot. Gesells. **24**: 1906.

[10] OSTENFELD, C. H., Experimental and cytological studies in the Hieracia. I. Castration and hybridization expermients with some species of Hieracia. Bot. Tidssk. **27**:225–248. *pl. 1.* 1906.

[11] REED, GEORGE M., Infection experiments with *Erysiphe graminis* DC. Trans. Wis. Acad. Sci. **15**:135–162. 1905.

Turgor in yeast.—PANTANELLI having examined the regulation of turgor in certain fungi and distinguished "cell pressure" into two factors, (1) osmotic pressure or turgor, due to solutes, and (2) tension, due to imbibition, has investigated by the same methods the turgor regulation in yeast derived from Roman bread.[12] He finds that during fermentation turgor at first increases, then remains constant, and finally diminishes when the nutritive value of the medium becomes much lowered. The power of osmotic regulation depends primarily upon nutrition. If the foods are temporarily removed without altering the concentration of the medium, the turgor and the tension diminish rapidly, the plant being compelled to use its own reserves, forming vacuoles. If grown in water or allowed to dry slightly, turgor diminishes, but the tension increases, in the first condition until death, in the second up to a maximum, after which it diminishes greatly before death. With age the power of osmotic regulation is gradually lost. Aeration facilitates it so much that it seems admissible to say that the Roman yeast lives during and after fermentation only because it falls into a state of narcosis more or less profound.—C. R. B.

Blast of rice.—METCALF[13] has recently published an account of the "blast" of rice with short notes on other rice diseases. This blast is characterized by lesions at one or more of the nodes of the stem above which the stem dies. It has often been confused with other diseases or injuries and the true extent of its damage not realized. The disease is promoted by resting the land or by applying nitrogenous fertilizers. It can be produced in healthy plants by inoculations directly from diseased plants, but the organism causing the disease has not yet been fully determined. The use of lime and marl with potash and phosphorus is recommended as fertilizer treatment that tends to reduce the tendency to blast. The disease is prevented by spraying with Bordeaux mixture, but this treatment is not practicable with this crop. The search for immune plants has been of no avail up to this time.—E. MEAD WILCOX.

A new chestnut disease.—MURRILL[14] has described a new and serious disease of the native chestnut, which is epidemic in many parts of New York City and threatens to destroy all the chestnut trees of that region. The disease is also known to occur in New Jersey, Maryland, District of Columbia, and Virginia. "The fungus attacks twigs, branches, and trunks, irrespective of size or position, and usually proceeds in a circle about the affected portion until it is completely girdled." It is described as a new species of Diaporthe (*D. parasitica*). —J. M. C.

---

[12] PANTANELLI, E., Richerche sul turgore delle cellule di lievito. Annali di Botanica 4: 1–47. 1906.

[13] METCALF, H., A preliminary report on the blast of rice, with notes on other rice diseases. Bull. N. Car. Exp. Sta. 121: 1–43. 1906.

[14] MURRILL, W. A., A new chestnut disease. Torreya **6**: 186–189. 1906.

Haustorium of Santalum.—BARBER[15] has begun the publication of a series of papers on root parasitism, the first one dealing with *Santalum album*, giving an account of the early stages of the haustorium as far as penetration into the host's tissues. This is a somewhat fuller account than that published by the author in the *Indian Forester* and noticed in this journal.[16] A second paper is promised which will describe the structure of the mature haustorium. Investigation of the mutual influence of host and parasite is also in progress.—J. M. C.

A new red clover.—BRAND[17] has published the results of a study of a red clover not hitherto used in the United States as a forage plant. The seed is from Orel, in the "Black Earth" region of Russia, and the plant possesses advantageous qualities that make its introduction desirable. Perhaps its most striking mark in the field is the almost complete lack of hairiness; but it is the general leafiness and the persistence and number of basal leaves that suggest for the new variety the name *T. pratense foliosum* Brand.—J. M. C.

Plant diseases in Nebraska.—HEALD[18] has published notes on the distribution and severity of numerous diseases of cultivated plants in Nebraska during 1905, together with suggestions as to treatment of the various troubles. This in a way constitutes a handbook of plant diseases in Nebraska. He has also published an account of a rot of apples due to *Sclerotinia fructigena*,[19] and a disease of the cottonwood due to *Elfvingia megaloma*,[20] once a member of the genus Polyporus.—E. MEAD WILCOX.

Pteridophytes of southern Florida.—EATON[21] has put on record his observations on the pteridophytes of southern Florida during three excursions. The purpose is to bring together the little-known species, with careful descriptions, so that they may be more readily recognized by students of the flora. The paper is also a contribution to geographical distribution. Most of the plants described are ferns, among which is a new species of Tectaria.—J. M. C.

---

[15] BARBER, C. A., Studies in root parasitism. The haustorium of *Santalum album*. 1. Early stages, up to penetration. Memoirs Depart. Agric. India **1**: no. 1. pp. 30. *pls.* 7. 1906.

[16] BOT. GAZETTE **40**: 159. 1905.

[17] BRAND, CHARLES J., A new type of red clover. U. S. Dept Agric., Bureau Pl. Ind., Bull. 95. pp. 45. *pls.* 3. 1906.

[18] HEALD, F. D., Report on the plant diseases prevalent in Nebraska during the season of 1905. Rep. Neb. Exp. Sta. **19**: 19–81. 1906.

[19] HEALD, F. D., The black rot of apples due to *Sclerotinia fructigena*. Rep. Neb. Exp. Sta. **19**: 82–91. *pls.* 1–2. 1906.

[20] HEALD, F. D., A disease of the cottonwood due to *Elfvingia megaloma*. Rep. Neb. Exp. Sta. **19**: 92–100. *pls.* 1–4. 1906.

[21] EATON, A. A., Pteridophytes observed during three excursions into southern Florida. Bull. Torr. Bot. Club **33**: 455–486. 1906.

Pollen tube of Houstonia.—MATHEWSON[22] has studied the advance of the pollen tube in Houstonia, coming to the conclusion that any mutual influence between the tube and the cells with which it comes in contact is very slight; and that the direction of the advance of the tube seems to be chiefly in response to a stimulus originating in the egg apparatus, perhaps in the egg itself.—J. M. C.

Operculina.—HOUSE[23] has published a synopsis of the genus Operculina as the second paper of his series on North American Convolvulaceae. It comprises about twenty tropical species, which are perhaps better known to many botanists under Convolvulus and Ipomoea. In North America fifteen species are recognized, two of them being described as new.—J. M. C.

Influence of temperature upon flowering of fruit trees.—Ecologists and physiologists will be interested in the phenological notes presented by SANDSTEN[24] relative to the influence of temperature and other factors upon the time of flowering of certain fruit trees. He reaches the conclusion that "physiological constants can be formulated from the climatic conditions during the ten months preceding the time of flowering."—E. MEAD WILCOX.

Effect of light on growth.—SELBY[25] has extended the work of MACDOUGAL to include four other species, mostly those which are latex-bearing. MACDOUGAL'S conclusions that light does not have a retarding influence on growth and that it does stimulate morphogenic processes in the meristematic tissues are confirmed.—RAYMOND H. POND.

Varieties of roots developed by English ivy.—Miss RANDOLPH[26] finds that *Hedera Helix* may be induced to form in all seven kinds of roots according to the conditions of moisture.—RAYMOND H. POND.

---

[22] MATHEWSON, C. A., The behavior of the pollen tube in *Houstonia coerulea*. Bull. Torr. Bot. Club **33**:487–493. *figs. 3.* 1906.

[23] HOUSE, H. D., Studies in the N. Am. Convolvulaceae. II. The genus Operculina. Bull. Torr. Bot. Club **33**:495–503. 1906.

[24] SANDSTEN, E. P., Conditions which affect the time of the annual flowering of fruit trees. Bull. Wisc. Exp. Sta. 137:1–21. 1906.

[25] SELBY, A. D., Studies in etiolation. Bull. Torr. Bot. Club **34**:67–75. *pls. 2. figs. 4.* 1906.

[26] RANDOLPH, HARRIET, The influence of moisture upon the formation of roots by cuttings of ivy. Bull. Torr. Bot. Club **34**:93–99. *figs. 4.* 1906.

NEWS.

DR. IRA D. CARDIFF, Columbia University, has been appointed professor of botany in the University of Utah, Salt Lake City.

DR. MAX KÖRNICKE, University of Bonn, has received the Buitenzorg grant of the German government for 1906–7 and left for this station August 29.

DR. C. A. J. A. OUDEMANS, the mycologist, emeritus professor of botany in the University of Amsterdam, died recently at Arnhem at the age of eighty years.

DR. D. H. CAMPBELL, Stanford University, has returned after an absence of a year, some of which was spent in South Africa and the Botanical Gardens at Peradeniya and Buitenzorg.

MR. LEROY ABRAMS, of the Smithsonian Institution, formerly instructor in botany in Leland Stanford University, has been appointed assistant professor of systematic botany in the University.

DR. BRADLEY M. DAVIS will spend next winter in Cambridge, Mass. His immediate work will be the completion with Mr. BERGEN of a laboratory and field manual to accompany the *Principles of Botany* which has recently appeared from the press of Ginn & Company.

THE GRANTS made for scientific research at the York meeting of the British Association include the following botanical grants: Physiology of heredity, £30; South African cycads, £35; Botanical photographs, £5; Structure of fossil plants, £5; Peat moss deposits, £7; Marsh vegetation, £15.

THE AUTUMN COURSE of public lectures announced by the Field Museum of Natural History, Chicago, contains two botanical titles as follows: October 20, "The century plants, and some other plants of the dry country," by Dr. WILLIAM TRELEASE; November 17, "Some phases of plant distribution," by Dr. J. M. GREENMAN.

COUNT OSWALD DE KERCHOVE DE DENTERGHEM, who upon the sudden death of Professor LÉO ERRERA was appointed president of the International Botanical Congress to be held in Brussels in 1910, died on March 20, at the age of sixty-two. He was president of the Royal Society of Agriculture, and of the Botanical Society of Gand, a senator, and ex-governor of Hainault.

AT THE University of Chicago, Dr. W. G. LAND has been promoted to an associateship, and Mr. L. L. BURLINGAME has been appointed assistant in morphology. Dr. FLORENCE LYON resigned at the close of the summer quarter, and shortly thereafter was married to Mr. S. VINCENT NORTON, of Akron, Ohio. The number of students registered for research work in the autumn quarter is the largest in the history of the department.

AT THE CONCLUDING MEETING of the International Conference on Hybridization and Plant Breeding the Veitch gold memorial medals were presented to Mr. W. BATESON, F.R.S., the president of the conference, Professor JOHANNSEN, Professor WITTMACK, and Professor MAURICE DE VILMORIN. Banksian silver-gilt medals were awarded to Miss E. R. SAUNDERS, lecturer on botany at Newnham College, and Mr. R. H. BIFFEN, for eminent services to scientific and practical horticulture.—*Nature.*

THE BOTANICAL SOCIETY OF AMERICA will meet in New York City in Convocation week, beginning December 27, in affiliation with the A. A. A. S. This meeting, the first after the union of the three constituent societies, will be an important one, as questions of future policy are likely to be brought before the society. The sessions will be arranged, in cooperation with the officers of Section G, so as to avoid conflict. Members are requested to send to the Secretary (WILLIAM TRELEASE, Missouri Botanical Garden, St. Louis), at the earliest possible date, titles of papers and information as to time and any special facilities required for their presentation. The total number of members at present is 119.

THE BOTANICAL SUBJECTS for 1907 and 1908 announced for the "Walker Prize," offered annually by the Boston Society of Natural History, are as follows:

For 1907: The structure and affinities of some fossil plant or group of fossil plants. The development of the gametophytes in any little known representative of the Coniferales. The anatomy and development of some order or group of the angiosperms.

For 1908: An experimental study of inheritance in animals or plants. A comparative study of the effects of close-breeding and cross-breeding in animals or plants. A physiological study of one (or several) species of plants with respect to leaf variation. Fertilization and related phenomena in a phenogamous plant. What proportion of a plant's seasonal growth is represented in the winter bud?